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MISE AU POINT D'UN OUTIL D'ÉVALUATION DE LA QUALITÉ BIOLOGIQUE DES PETITES RIVIÈRES DE GUYANE SUR LA BASE DES INVERTÉBRÉS BENTHIQUES

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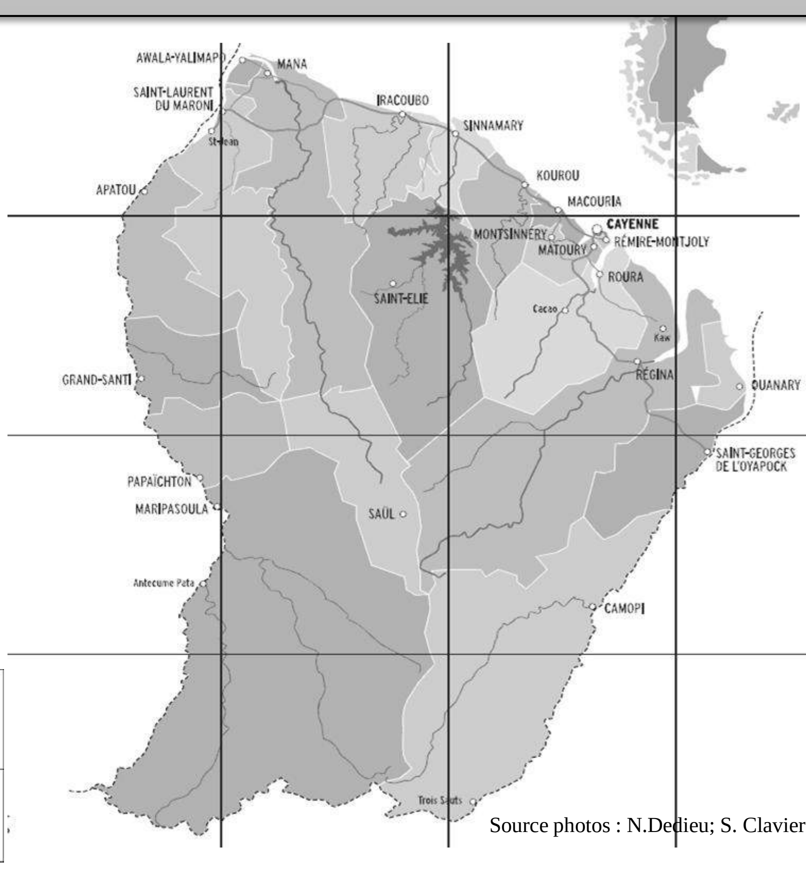
Mise au point d'un outil d'évaluation de la qualité biologique des petites rivières de Guyane sur la base des invertébrés aquatiques

Nicolas Dedieu, 2014



Source photos : N.Dedieu; S. Clavier; *Monfort et Ruf, 2005*

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Résumé

La Directive Cadre Européenne sur l’Eau (DCE) a imposé à chaque état membre l’objectif d’obtenir une amélioration de la qualité des eaux de surface, pour d’atteindre le bon état écologique à l’horizon 2015. Cette directive implique non seulement d’évaluer le degré de pollution mais aussi de définir l’état écologique des milieux par rapport à un référentiel (se rapprochant de l’état « naturel »). Cela a conduit à définir une typologie des rivières basées sur les «conditions de référence». Bien que géographiquement éloignés du continent, les DOM font partie intégrante de l'Union européenne et sont soumis aux mêmes objectifs et obligations en termes de politiques environnementales. La Guyane française (GF) est une région d'outre-mer de la France située sur la côte nord-est de l'Amérique du Sud. La forêt primaire guyanaise reste l'une des moins touchés du monde, cependant, les exploitations aurifères et sylvicoles ont de fortes répercussions sur les milieux aquatiques. Les petits cours d’eau (profondeur inférieur à 1 m et da largeur inférieur à 10 m) représentent 70-80% du réseau hydrographique de la GF. La plupart des petits cours d'eau sont situés dans les zones boisées et présentent haute qualité écologique et certains, sinon la plupart d'entre eux, n'ont jamais été touchés par une activité humaine. L'objectif principal de cette thèse était de concevoir un indice capable d'évaluer la qualité écologique des petits cours d'eau de la GF.

Nous avons d’abord analysé l’effet des perturbations humaines sur la qualité physicochimique des écosystèmes. Puis, nous avons établi la première typologie de la structure des communautés benthiques en fonction des conditions environnementales. L'identification de ces sous-régions écologique et des conditions de références nous a permis de construire un indice multimétrique. Enfin, une approche fonctionnelle basée sur les Ephémères a été utilisée afin de fournir de nouvelles informations sur l'écologie de cet ordre.

Nous avons démontré que les variables physiques relatives au fond des cours d’eau et les matières en suspension différencient les sites orpaillés, tandis que les concentrations en nutriments ne sont pas significativement modifiées par les impacts humains. La biotypologie a montré que les sites caractérisés par des communautés d'invertébrés sont regroupées en deux sous-régions principales: la plaine alluviale

côtière et le plateau des Guyanes. Les changements dans la composition de la communauté, et à un degré moindre la richesse taxonomique au sein de chaque sous-région ont révélé des impacts écologiques liés à l'exploitation aurifère et forestière. Parmi les mesures décrivant la structure des communautés (métriques) calculées à partir des deux sous-ensembles de sites de références et impactés, nous avons sélectionné les paramètres présentant le meilleur compromis entre forte efficacité de la discrimination, faible spécificité, faible redondance et une grande stabilité dans les conditions de référence. L'IBMG est composé de deux métriques taxonomiques de richesse, deux métriques basées sur l'abondance, une métrique relative au trait biologique et un indice de diversité. Les résultats sur les communautés d'Ephémère ont mis en évidence le rôle important de cet ordre pour la bioévaluation de petits cours d'eau soumis à l'exploitation aurifère. Des recherches futures fondamentales sur la taxonomie et l'écologie de cet ordre pourront permettre d'améliorer l'outil d'évaluation biologique actuel. Enfin, ce travail à souligné le fait que les petites masses d'eaux de tête de bassin de Guyane abritent potentiellement une faune riche et spécifique.

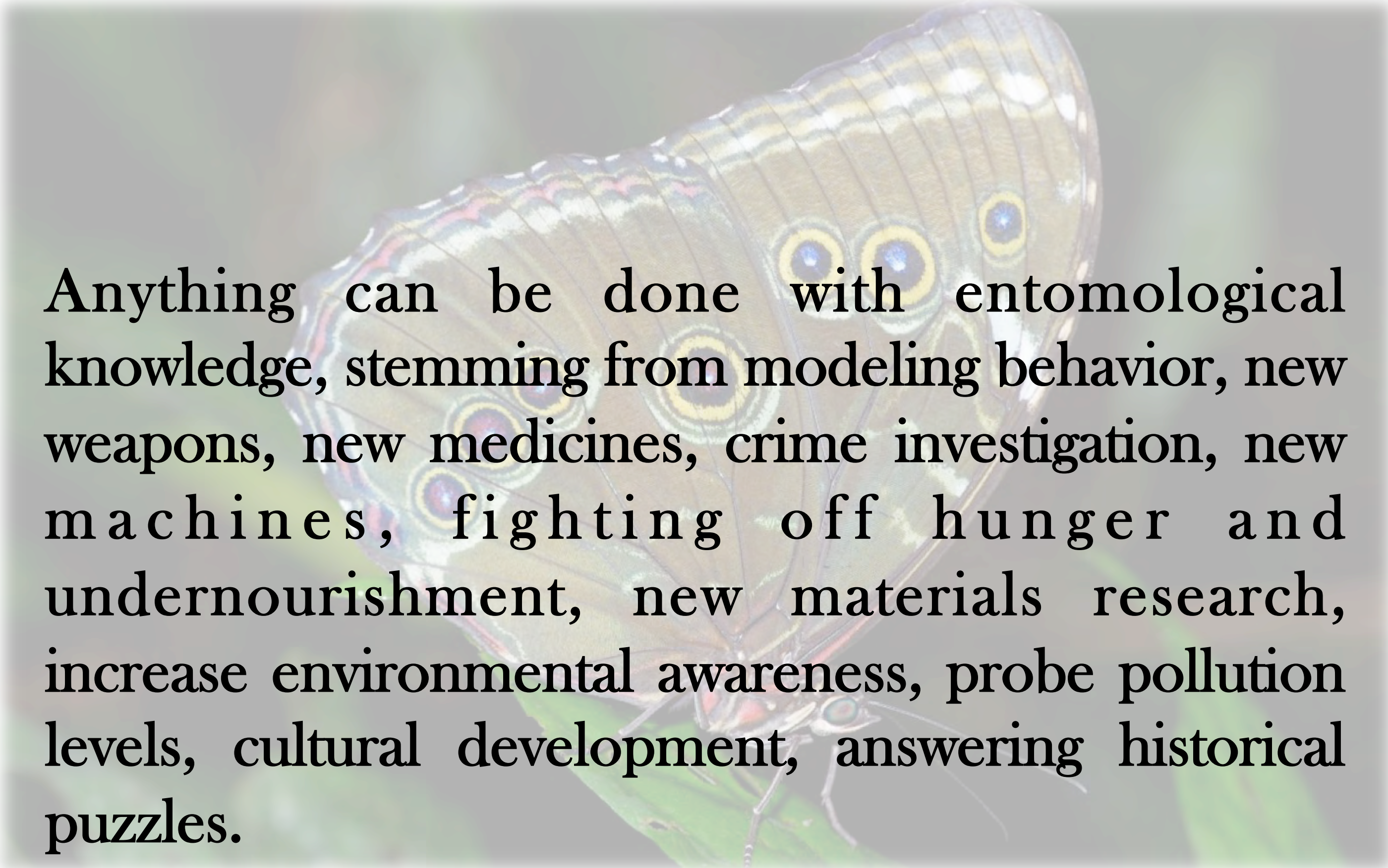
Abstract

The European Water Framework Directive (WFD) aims to achieve a rapid improvement of the water quality, in each member state, and to achieve good ecological status of rivers by 2015. This new directive implies not only to assess the degree of pollution but also to define an ecological state of the environment in relation to a reference condition. This has prompted a large amount of works which yielded characterizations of both reference physical–chemical environments and biological communities in continental Europe, as well as practical tools (e.g., biological indices) to evaluate water quality. Although geographically distant the continent, DOMs are an integral part of the European Union and are subject to the same objectives and obligations in terms of environmental policies. French Guiana (FG) is an overseas region of France located on the north-eastern coast of South America. The Guianese primary forest remains one of the least impacted of the World, however, gold mining and timber have strong localized impacts upon river ecosystems. Small streams (from headwaters to rivers with depth inferior to 1 meter and width inferior to10 meters) represent 70–80% of all running waters in FG. Most small streams are located in forested areas and exhibit high ecological quality; some if not most of them have never been impacted by any human activity.

The main objective of this thesis was to design a biotic index to assess the ecological quality of small streams of FG. We first analysed the effects of disturbance on the physical-chemical quality of ecosystems. Then, we brought out the first typology of community structure in relation to environmental conditions. The identification of the sub-ecological regions and the references conditions allowed us to build a first multi-metric. Finally, a functional approach based on Ephemeroptera was used in order to provide new information on the ecology of this important order of macroinvertebrates.

We first demonstrated that physical variables describing the river bottom, and suspended solids discriminate gold-mined from reference sites, while, whatever the type of impact encountered, nutrient concentrations do not prove relevant to measure human impacts. The bio-typology showed that sites characterized by invertebrate communities clustered into two major subsets: the coastal alluvial plain and the Guiana Shield. Changes in community composition, and to a lesser extent taxonomic

richness within each sub region revealed ecological impacts of gold mining and logging. Among the biological metrics calculated from the hydro-ecoregions subsets of reference and impaired sites, we selected metrics exhibiting the best trade-off between high discrimination efficiency, low specificity, low redundancy and high stability under reference conditions. The IBMG is composed of two taxonomic richness-based metrics, two abundance-based metrics, one trait-related metric and a diversity index. Results on Ephemeroptera communities put in light the important role of this order for the bio-evaluation of small streams submitted to gold-mining. Future fundamental research on the ecology and the taxonomy of this order should thus improve the current bioassessment tool. Finally, this work highlighted the fact that headwater stream small of FG potentially harbor a rich and specific fauna.



Anything can be done with entomological knowledge, stemming from modeling behavior, new weapons, new medicines, crime investigation, new machines, fighting off hunger and undernourishment, new materials research, increase environmental awareness, probe pollution levels, cultural development, answering historical puzzles.

As insects are almost everywhere and all the time, and they are diverse beyond comprehension and so utterly complex, there is always an application for whatever you may think of.

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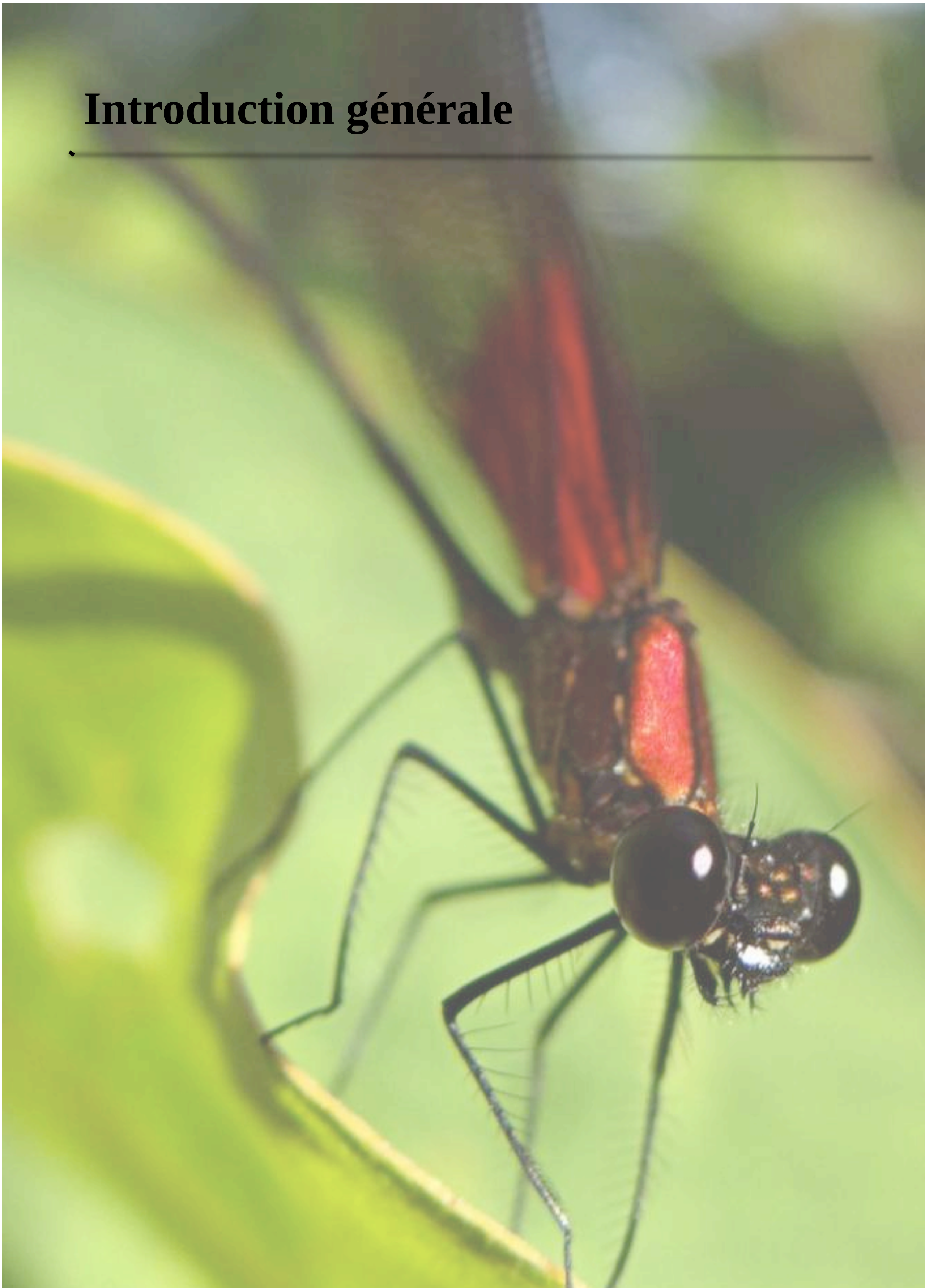
Introduction générale



En 1957, Hutchinson définit la **niche écologique** comme un hypervolume où chaque dimension de l'espace représente une ressource (ex.: alimentaire, habitat) ou une condition de l'environnement (température, précipitations). Avec la quantité de ressources qui varie dans l'espace et dans le temps, ces paramètres seraient donc des conditions limitantes que l'on peut hiérarchiser pour étudier la vulnérabilité d'une espèce dans son environnement. Chaque espèce serait donc capable de tolérer, pour un ensemble de facteurs écologiques, un sous-ensemble des conditions possibles. Il est alors possible, lorsque l'on considère *n* facteurs écologiques pouvant influencer la survie d'une espèce, de définir un volume à *n* dimensions représentant les conditions dans lesquelles l'espèce est capable de survivre. Ce volume traduirait donc les exigences environnementales de chaque espèce (la niche fondamentale ou potentielle - Grinnell, 1917 ; Elton, 1927). Prenons l'exemple de deux espèces A et B qui présentent des niches écologiques fondamentales se recouvrant partiellement (Figure 1).

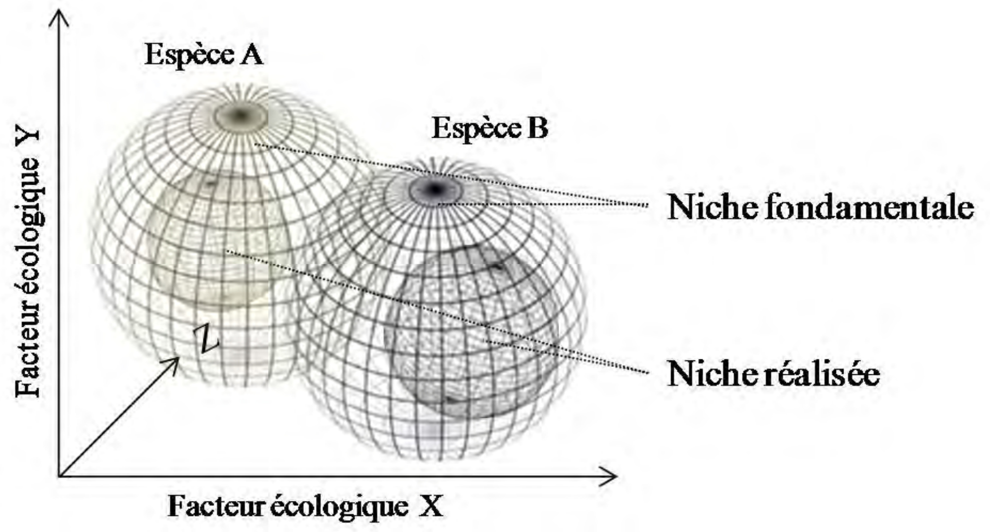


Figure 1: Représentation schématique des niches écologiques fondamentales et réalisées de deux espèces (A et B) dans un espace défini par trois facteurs écologiques (x, y et z) (Hutchinson, 1957).

La théorie de la différenciation de niche prédit que des espèces ayant des niches écologiques trop similaires ne peuvent coexister (Gause, 1934). Cette différenciation traduit un processus selon lequel, la sélection naturelle va conduire les espèces concurrentes à utiliser des ressources ou des niches différentes. La niche écologique réalisée de chacune de ces espèces correspondrait alors à un sous-ensemble de chacune de leurs niches fondamentales où les espèces seraient effectivement présentes limitant ainsi la compétition entre les espèces avec des niches similaires (MacArthur et Levins, 1967). La notion de niche écologique a été pendant longtemps presque exclusivement associée à la notion de compétition, a ainsi progressivement été remise en question dans les mécanismes de structuration des communautés (Chase et Leibold, 2003). Cette remise en question a été initiée par la **théorie unifiée neutraliste de Hubbell** (2001) qui exprime que tous les individus d'une communauté ont la probabilité identique de se reproduire, migrer ou mourir (Figure 2). Cette théorie, qui a démontré une capacité surprenante à rendre compte de l'abondance relative des espèces ainsi que de leur assemblage spatial, a permis de raviver l'intérêt des écologistes des communautés pour la théorie de la niche écologique et d'inciter à la réflexion sur la redéfinition de son concept (Chase et Leibold, 2003).

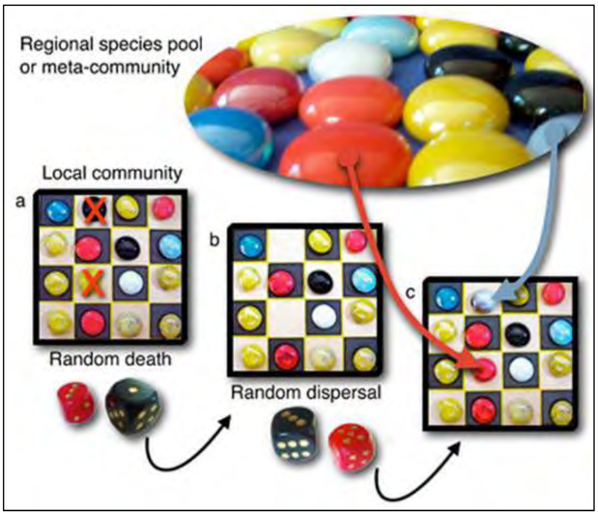


Figure 2: Exemple d'un processus neutre. Une communauté locale (a) correspond à un sous-ensemble de toutes les espèces présentes dans la région. Certains individus vont mourir au hasard au sein de cette communauté locale et créer des « sites ouverts » (b). Ces Sites ouverts vont ensuite être remplis par un processus de dispersion aléatoire à la fois au sein de la méta-communauté et de la communauté locale (c). Cette séquence va ainsi se répéter et provoquer un phénomène de dérive écologique où l'abondance des espèces va changer de manière aléatoire au cours du temps (source : Harpole, 2010).

En 2006, McGill et al. soulignent que la compétition n'est pas l'unique facteur structurant des communautés mais que les gradients environnementaux ont également un rôle important sur la structuration des communautés. En effet, l'étude des facteurs contrôlant l'agrégation des populations en une communauté est un point primordial en écologie des communautés et est souvent abordé en utilisant la notion de **règles d'assemblage** (Keddy, 1992a ; Weiher et Keddy, 1995 - Figure 3). Cette approche permet entre d'autre d'aborder une question fondamentale qui est de savoir *comment se structurent les communautés locales à partir d'un grand pool régional d'espèces*. Le principe de ces règles d'assemblages va permettre de donner une série de critères permettant de prédire, à partir d'un ensemble régional d'espèces, la combinaison d'espèce qui formera une communauté au niveau local, dans un habitat spécifique (Keddy, 1992a). Cette structuration des communautés locales se ferait au travers d'un filtre environnemental qui s'organiserait de façon hiérarchique.

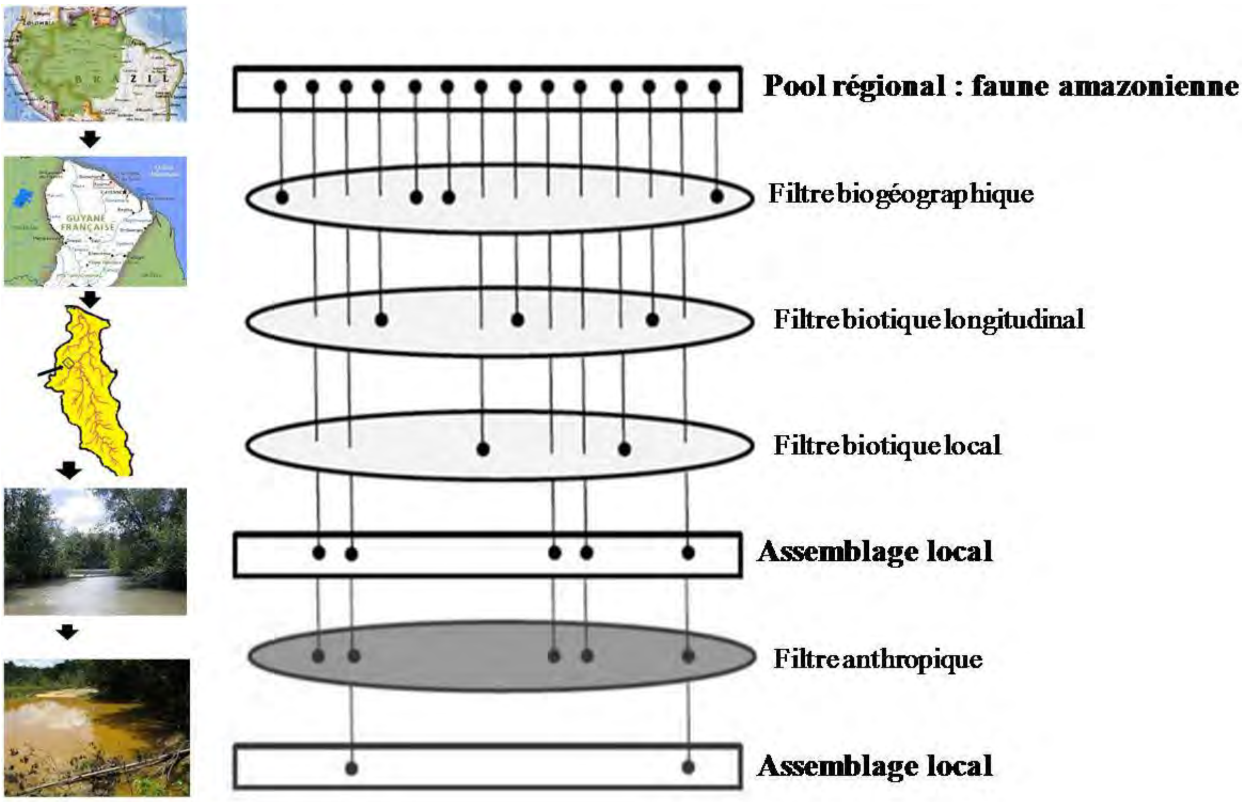


Figure 3 : La structuration des communautés (Schéma modifié de Keddy, 1992a)

La composition d'un assemblage n'est pas aléatoire et résulte d'une série de filtres hiérarchiques agissant à des échelles temporelles et spatiales différentes (Tonn, 1990; Keddy, 1992b, Jackson et al., 2001). En effet, les assemblages locaux sont déterminés non seulement par des mécanismes locaux et contemporains, mais aussi par des processus agissant à des échelles spatiales et temporelles plus larges (Figure 3) :

- A large échelle agit un filtre biogéographique et historique. Il est défini par les événements climatiques majeurs que sont les périodes glaciaires et interglaciaires, et par les événements géologiques qui ont façonné la topographie du territoire. Ces événements ont façonné un pool d'espèces homogène qui correspondant au pool régional Amazonien.
- Au sein du pool d'espèce régional agit un nouveau filtre définissant la faune potentiellement présente à l'échelle du tronçon de cours d'eau. Il s'agit ici d'un ensemble de contraintes écologiques non aléatoires qui évoluent progressivement le long du gradient amont aval d'un cours d'eau, et forment un continuum (ex: Vannote et al., 1980). Les conditions physiques notamment, évoluent graduellement. La largeur et profondeur du cours d'eau, la distance à la source, la pente et l'altitude sont communément utilisées pour se situer au sein

de ce gradient (Jackson et al., 2001). Les espèces qui possèdent les traits permettant de s'adapter et de survivre aux contraintes qui sont présentes localement peuvent s'établir (Keddy, 1992b). Les espèces ne pourront s'établir sur l'ensemble du gradient, car leurs capacités physiques et physiologiques ne leur permettant pas de supporter l'ensemble des conditions environnementales du gradient.

- A une échelle encore plus fine (habitat), s'exerce un dernier filtre, dit « local », correspondant aux contraintes abiotiques (et biotiques) agissant localement. Ce dernier filtre correspond à la complexité structurelle de l'environnement. La structure de la communauté locale dépendraient alors des caractéristiques de leur habitat : introduit par la théorie de l'« Habitat Templet » (Southwood, 1977 ; 1988) adaptée quelques années plus tard aux cours d'eau (« River Habitat Templet » ou RHT : Townsend et Hildrew, 1994). Cette théorie est basée sur le fait que l'habitat, et plus particulièrement sa variabilité spatio-temporelle, constitue le patron dans lequel les stratégies d'histoire de vie des organismes sont forgées. Les organismes seraient plus sélectionnés en fonction de leurs capacités de résistance et de résilience aux perturbations (naturelles ou humaines) qu'en fonction de leurs aptitudes compétitrices. Les caractéristiques structurantes de l'habitat seraient, du point de vue des organismes, (i) la fréquence, l'intensité et la prédictibilité des perturbations (Southwood, 1977; Townsend et Hildrew, 1994) et (ii) l'hétérogénéité spatiale qui, par la mise à disposition de zones refuges, permettrait d'améliorer les chances de survie des organismes aux perturbations (Townsend, 1989 ; Townsend et Hildrew, 1994). Les contraintes biotiques (compétition, prédation) seraient indirectement prises en compte car les pressions de compétition et de prédation s'exerçant dans un milieu sont dépendantes de la structure de l'habitat local (Jackson et al., 2001). La prise en compte de l'ensemble des filtres, via la mesure de variables environnementales représentant les différents filtres hiérarchiques permet donc de faire la prédiction de présence ou d'absence de chaque espèce dans un habitat considéré.
- La modification de l'environnement par les activités humaines (filtre anthropique), va ensuite potentiellement impacter la composition d'un assemblage local car certaines espèces ne pourront pas supporter les nouvelles conditions environnementales. C'est sur le principe que repose la bioindication (Wright et al., 2000; Carter et Resh, 2001; Niemi et McDonald, 2004).

Un **bioindicateur** est un indicateur végétal, animal ou un groupe d'espèces dont la présence ou l'état renseigne sur certaines caractéristiques écologiques de l'environnement. Le principe est d'observer des effets biologiques ou éco-systémiques, au niveau de l'individu et/ou de populations ou écosystèmes. L'utilisation des **communautés** biologiques pour l'évaluation de la qualité environnementale est une approche très répandue car elle est pertinente pour mettre en évidence l'impact des activités anthropiques sur les écosystèmes (Warwick, 1993, Clements et Kiffney, 1994 ; Attrril et Depledge, 1997). Les communautés biologiques sont de bon intégrateur de leur milieu car elles reflètent les caractéristiques et la variabilité spatiotemporelle de leur habitat, qu'il soit modifié ou non par les activités anthropiques (Gorman et Karr, 1978; Townsend, 1989 ; Townsend et Hildrew, 1994 ; Usseglio-Polatera, 1997 ; Stevenson et Pan, 2010). Les premières méthodes de bioindication ont fait leur apparition dans la littérature scientifique européenne dès le début du 20e siècle avec par la méthode dite des **« saprobies »** (Kolkwitz et Marsson, 1909), basée sur la description des exigences écologiques des espèces vis-à-vis de la pollution organique. En France, le véritable essor de la bioindication a eu lieu dans les années 70, avec la mise au point du premier indice basé sur les macroinvertébrés benthiques (l'indice biotique IBG - Verneaux et Tuffery, 1967) qui a abouti à l'indice de qualité biologique global (IQBG) en 1976, puis à l'indice biologique global (IBG) en 1985. Cet indice a ensuite été normalisé en 1992 sous forme d'indice biologique global normalisé (IBGN). Au niveau international, les années 80 ont vu l'avènement de la **notion d'intégrité biotique** et le développement des premiers outils de bioindication dits « **multimétriques** » (c'est-à-dire basés sur plusieurs caractéristiques taxonomiques ou fonctionnelles d'un compartiment biologique considérées simultanément). Ce type d'approche a été proposé en premier lieu par Karr (1981; Karr et al., 1986) pour les communautés de poissons de cours d'eau nord-américains. Ce concept est défini comme « *la capacité de supporter et maintenir une communauté d'organismes équilibrée et intégrée, ayant une composition spécifique, une diversité et une organisation fonctionnelle comparables à celles d'habitats naturels de la même région*». Ces travaux fondateurs ont donné lieu au développement d'un premier indice multimétrique pour les poissons en France, l'indice poissons rivière (IPR ; Oberdorff et al., 2001, 2002). L'application d'indices multimétriques s'est progressivement étendue à d'autres organismes biologiques, notamment les invertébrés benthiques qui sont progressivement devenus des outils majeurs de la biosurveillance des milieux aquatiques dans différents pays européens ainsi qu'aux Etats-Unis (Rosenberg et Resh, 1993; Chessman, 1999; Reynoldson et al., 2001; Resh et Rosenberg, 2008 Wright et al., 1998 (RIVPACS); Hering et al., 2003 (AQEM Project) ; Furse et al., 2006 (STAR Project). Ces organismes dont la taille est supérieure à 3-5mm au dernier stade de leur développement et vivant dans la colonne d'eau et dans les premiers centimètres de la couche de sédiments sont de parfaits intégrateurs de la qualité globale du milieu aquatique. De plus, ils présentent des caractéristiques intéressantes pour la conception d'outils biologiques (Archaimbault, 2003) qui sont:

- une large répartition géographique (rendant les méthodes comparables à grande échelle),
- une durée de vie relativement longue (quelques mois à quelques années),
- une sédentarité au sein de leur habitat,
- Une grande diversité de forme due au grand nombre de taxons et de phyla et appartement à ce groupe. Cette grande hétérogénéité leur permet de couvrir un large spectre de réponses aux perturbations (Rosenberg et Resh, 1993).

Les outils biologiques permettent de renseigner **l'état écologique** d'une masse d'eau et donnent une information quantitative sur la structure et du fonctionnement des écosystèmes aquatiques (espèces végétales et animales, hydro-morphologie et physico-chimie). Sur le plan opérationnel, il y a deux objectifs principaux dans l'établissement d'indices biotiques pour le contrôle de la qualité des cours d'eau :

- Le premier est le contrôle de l'intégrité des milieux aquatiques sur un territoire. Il consiste de détecter des perturbations dans des milieux «inconnus» de façon ponctuelle. C'est l'aspect **spatial** du contrôle (ex. : étude d'impact, état des lieux),
- Le deuxième est le suivi dans des points fixes de la qualité du milieu aquatique. C'est l'aspect **temporel** du contrôle. C'est le cas du **réseau de contrôle de surveillance (RCS)** qui permet d'évaluer l'état général des eaux et les tendances d'évolution au niveau d'un bassin,

En 2003, la Directive Cadre Européenne sur L'eau (DCE - European Commission, 2003) a fixé des objectifs ambitieux pour la préservation et la restauration de l'état des eaux superficielles et souterraines : **l'atteinte d'ici 2015 du « bon état » de l'ensemble des masses d'eau européenne.** Cet objectif n'est atteint qu'à la double condition de justifier d'un **bon état chimique** et d'un **bon état écologique** (Figure 4).

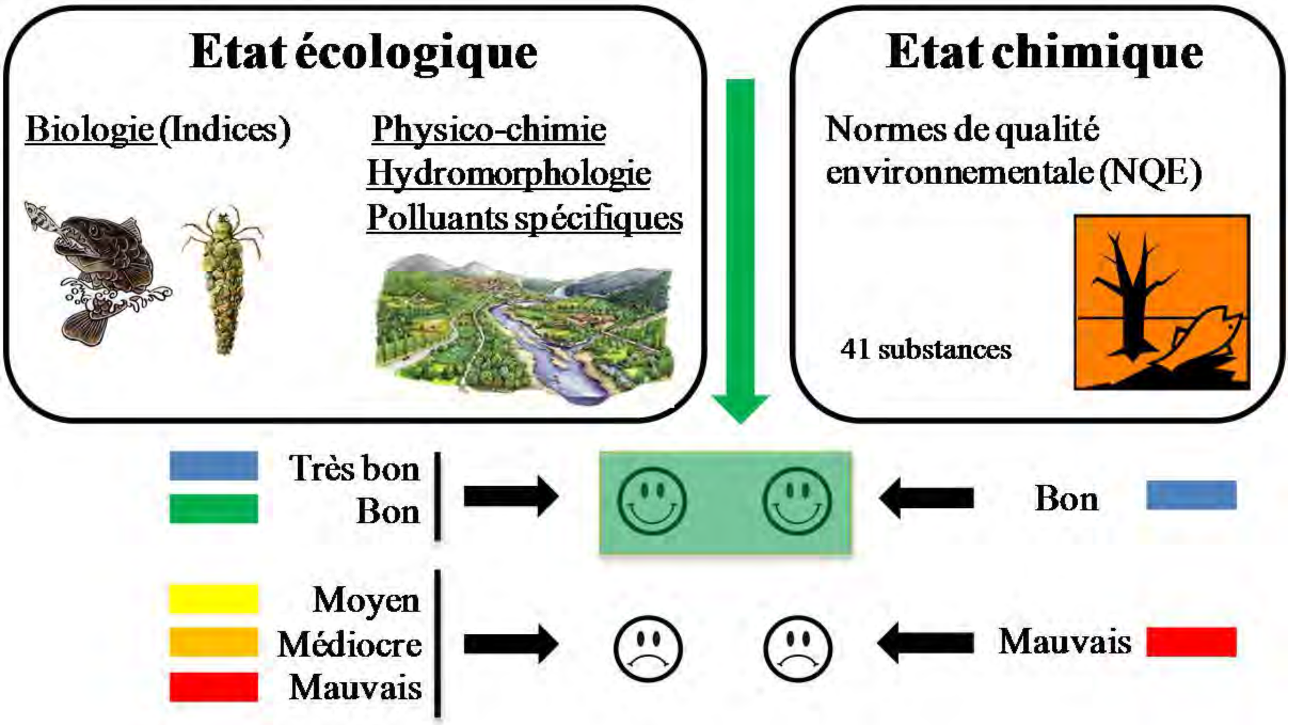


Figure 4: Principe de l'évaluation du « bon état » d'une masse d'eau selon la DCE.

Satisfaire cette demande nécessite de disposer d'indicateurs biologiques capables d'apporter une information pertinente sur l'état de santé de ces écosystèmes. La DCE impose aux États membres des exigences radicalement nouvelles en termes de prise en compte des différents compartiments biologiques et de fiabilité à atteindre dans l'évaluation de l'état écologique. L'annexe V de la directive demande en particulier aux États membres de mettre au point, **pour chaque catégorie de masses d'eau superficielle**, un outil de bioindication **pour chacun des éléments de la qualité biologique** (exemple : poisson, macroinvertébrés, diatomées). Ces exigences se sont traduites par un effort sans précédent pour le développement de méthodes de bioindication. Pour les organismes de recherche chargés de la mise au point de ces méthodes d'évaluation, l'enjeu est double : il s'agit de construire, à partir de rien ou sur la base de méthodes préexistantes, **des outils de bioindication pertinents** au plan scientifique et adaptés à une mise en œuvre à grande échelle, mais aussi d'assurer la **compatibilité de ces méthodes** avec le cadre précis imposé par la DCE. La nécessité, mise en avant par la DCE d'évaluer l'état écologique du milieu dans une perspective intégrée, c'est-à dire reflétant l'ensemble des impacts biologiques liés aux pressions pesant sur un milieu, a entrainé le plus souvent à la construction d'indices dits **multimétriques** (Hering et al., 2006, Lücke et Johnson, 2009). Ces indices sont des combinaisons de plusieurs métriques pondérées selon certains critères, qui ensemble, sont présumées représenter une gamme de réponses des communautés biologiques aux perturbations d'origines anthropiques.

Cette Directive implique également d'évaluer le degré de pollution par rapport à un référentiel (se rapprochant de l'état « naturel »). Cela a conduit à définir une **typologie des rivières basées sur les «conditions de référence**» (Reynoldson et al., 1997) exprimés en **« ratio de qualité écologique » (Ecological Quality Ratio - EQR)** permettant de rendre compte d'un écart à la référence (Hering et al., 2006). Ces indicateurs doivent être scientifiquement valides, efficaces, rapides à mettre en œuvre et applicables à différentes régions géographiques.

Membre de L'union Européenne, la France comprend 5 départements **d'Outre-mer** (DOMs) qui sont : la Guyane Française, la Martinique, la Guadeloupe, la Réunion et Mayotte. Bien qu'étant géographiquement éloignés du continent, les DOMs font partie intégrante de l'union Européenne et sont donc soumis aux mêmes objectifs et obligations en termes de politique environnementale. Par conséquent, la Directive Cadre Européenne sur l'Eau (DCE - European Council, 2000) doit également être appliquée aux DOMs bien que ces zones accusent d'un fort retard dans la gestion de la politique de l'eau. Pour les besoins de la DCE, tout comme la France métropolitaine, les cours d'eau de Guyane ont été découpés en **masses d'eau**, qui correspondent à des tronçons homogènes de cours d'eau. En 2006, un premier état des lieux a estimé que le réseau hydrographique de Guyane était extrêmement dense et ramifié. Suite à la publication du référentiel hydrographique BD Carthage® en 2013 (Mourguiart et Linares, 2013) et au schéma directeur d'aménagement et de gestion des eaux du district de la Guyane -2015, le district de la Guyane a été découpé en 841 masses d'eau de surface. Le référentiel BD Carthage® répertorie 112000 km de cours d'eau répartis sur 8 grands bassins versants (Mourguiart and Linares, 2013) dont 80 % du réseau hydrographique correspond à des rivières de moins de 10 m de large et moins d'un mètre de profondeur moyenne, dénommés : « cours d'eau de tête de bassin » ou « petite masses d'eau » (PME). Cependant, avant la mise en œuvre des travaux décrits dans cette thèse**, aucun réseau de surveillance, ni indicateur n'existait pour les PME.** Les connaissances sur ces milieux particuliers et reculés sont fragmentaires en Guyane, tant en ce qui concerne leur fonctionnement écologique ainsi que la faune qui y vit. Les cours d'eau de tête de bassin sont caractérisés par des interactions très fortes et dynamiques avec les systèmes qui les entourent. Les apports allochtones et les matières particulaires, par exemple, jouent un rôle central dans la détermination des conditions physiques et chimiques des cours d'eau (Likens et Bormann, 1974) ainsi que dans la régulation de la composition et la productivité des communautés (Wallace et al., 1997). En raison de cette étroite relation terrestre/aquatique, les cours d'eau de tête de bassins et les espèces qu'ils abritent sont très sensible aux perturbations naturelles et anthropiques. Mettre en place des méthodes d'évaluation et de suivi pour ces masses d'eau est hautement nécessaire car ces petites masses d'eau correspondent à l’empreinte chimique de l'ensemble du réseau hydrographique. De plus, du fait que l’activité humaine en Guyane continue de s'étendre vers l’intérieur des terres, l’effet cumulatif des perturbations en tête de bassins affectera probablement les ressources en aval de façon croissante (Lowe et Likens, 2005).

Le concept général porté par la DCE peut virtuellement être appliqué partout dans le monde : la typologie des cours d’eau et l’étalonnage à partir de conditions naturelles qui représentent les principes fondateurs pour l’application de la DCE ne posent pas de contrainte particulière en Guyane. Par conséquent, le développement d’un outil d’évaluation DCE-compatible pour les PME de Guyane semble donc réalisable. Une première acquisition des données puis une validation des méthodes au sens de la DCE seront nécessaires pour valider ce travail. Pour répondre à ces objectifs, nous sommes donc confrontés à deux problématiques majeures:

- **La collecte des données**: Les données relatives aux invertébrés des cours d’eau et aux variables de la qualité environnementale ont été collectées depuis quelques années en Guyane. Cependant, l’un des problèmes majeurs pour l’analyse de ces informations reste la faible homogénéité des données collectées. Un effort d’échantillonnage important sur l’ensemble du territoire a ainsi été entrepris. Ce jeu de données a permis d’obtenir une représentation aussi fiable que possible du territoire et des habitats disponibles, aussi bien dans les zones de référence que dans des sites soumis à des pressions anthropiques.
- **Le besoin d’adapter les méthodes de construction d’un indice aux spécificités de la faune, des habitats et des perturbations rencontrées en Guyane**. La définition des conditions écologiques de référence, l’expression des résultats en termes d’écart à la référence (EQR), la création d’une échelle de qualité traduisant la santé des écosystèmes et l’impact des perturbations anthropiques sur la faune.

A ce jour, des indices DCE-compatibles ont été élaborés pour la France (I2M2 - Mondy et al., 2012), la Réunion (IRM – Picot, 2012), les Antilles Françaises (IBMA – Touron-Poncet et al., 2014). Or, ces indices qui ont été mis au point pour des rivières continentales Européennes ou dans des milieux insulaires néotropicaux ne peuvent être transposés à la **Guyane Française** ou les contextes biogéographiques et climatiques d’une part, et le retard de connaissance de la faune d’autre part, ne permettent pas d’appliquer ces protocoles préexistants. En ce qui concerne l’application de la DCE pour les invertébrés et les sujets liés, la situation actuelle met en évidence l’absence d’application d’une typologie des cours d’eau. Une proposition de typologie basée sur le travail de Chandesris et al. (2005) est précisée en prenant en compte les communautés d'invertébrés aquatiques. Afin de construire une outil biologique DCE-compatible et donc spécifique à la Guyane, nous nous sommes basés sur de récents travaux qui prennent en compte les variations naturelles inter et/ou intra-régional de la structure des communautés (Oberdorff et al., 2002; Pont et al., 2006 ; Mondy et al., 2012). Ces nouveaux indices sont basés sur des modèles prédictifs qui permettent d'évaluer les prédictions spécifiques au **niveau d'un site** en l'absence d'impact anthropique et indépendamment des facteurs environnementaux naturels. Cette approche passe par un ajustement des paramètres biologiques (métriques) pour prendre en compte la variabilité naturelle avant d'analyser leurs réponses aux perturbations anthropiques (Figure 5).

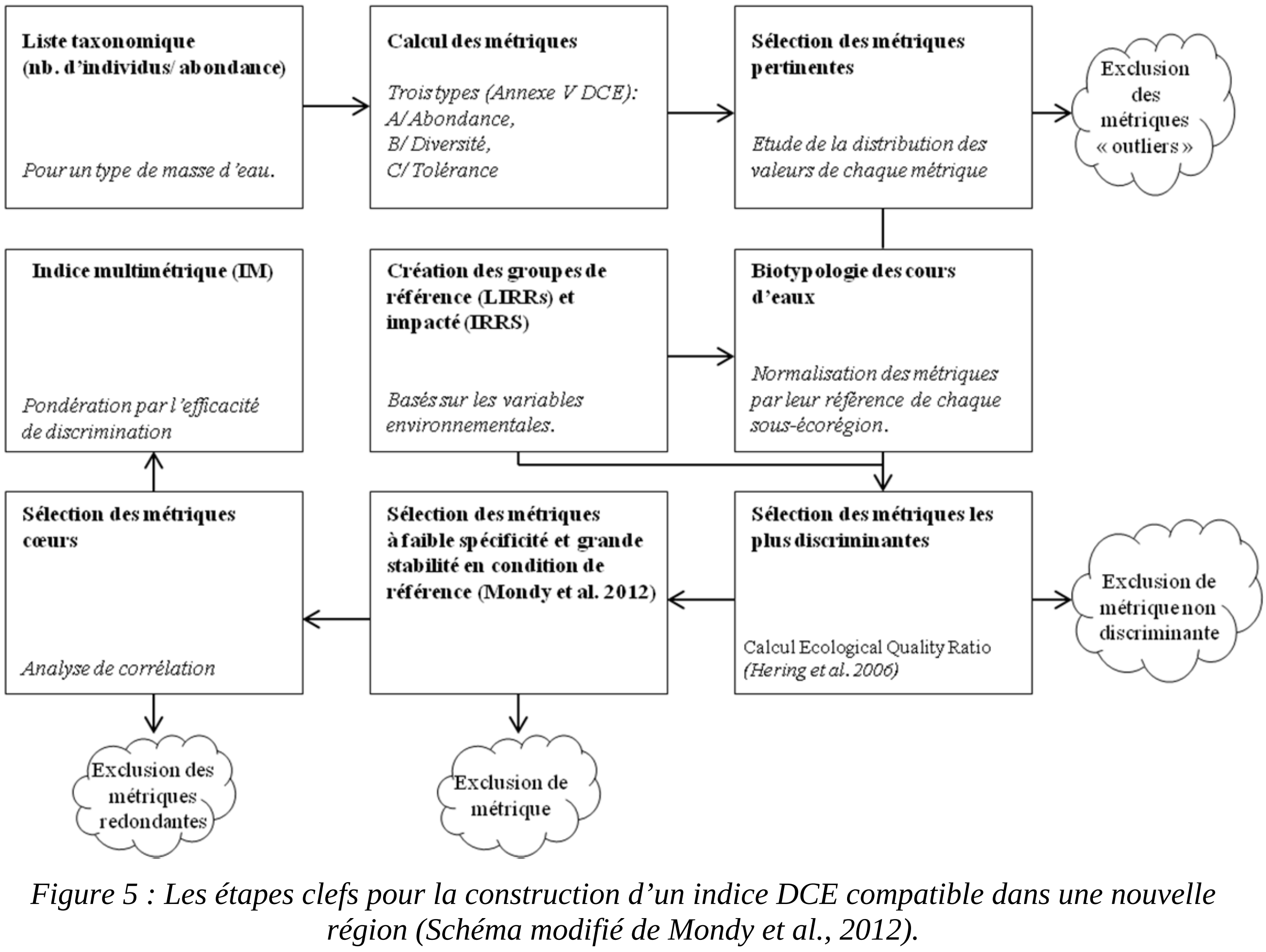


Figure 5 : Les étapes clefs pour la construction d'un indice DCE compatible dans une nouvelle région (Schéma modifié de Mondy et al., 2012).

Ce travail de thèse s'articule en six chapitres.

Le premier chapitre décrit de façon plus détaillée la zone d'étude, les caractéristiques écologiques des petites masses d'eau ainsi que les perturbations anthropiques typiques de la Guyane. Ce chapitre nous a semblé nécessaire à une bonne compréhension des systèmes étudiés et de leur contexte écologique / économique plus vaste, dans la mesure où les chapitres suivants se présentent sous la forme de manuscrits publiés ou en voie de publication.

Le second chapitre pose la question fondamentale de l'effet des principaux types de perturbation rencontrés en Guyane sur la physico-chimie des petits cours d'eau de tête de bassin. Les principales perturbations sont le fait de l'activité d'orpaillage et de l'exploitation forestière. Nous pouvons donc nous attendre à des effets essentiellement physiques sur les cours d'eau, et nous avons émis l'hypothèse selon laquelle l'orpaillage affecte directement le lit des rivières à un effet très supérieur à celui de la déforestation qui affecte le milieu riverain. Enfin, en condition non impactée, nous avons vérifié l'hypothèse selon laquelle la chimie de l'eau reste relativement homogène.

Le troisième chapitre présente la classification des petits cours d'eau de Guyane basée sur les communautés de macroinvertébrés. Cette étape, bien que basée sur une description des communautés et des environnements physico-chimiques associés, est nécessaire pour la mise en place d'un indice biotique basé sur l'écart à la référence car elle teste l'hypothèse selon laquelle la structure des communautés permet de délimiter des sous-régions ou hydro-écorégions écologiquement comparables en conditions de référence, tout en s'assurant que pour chaque ensemble ainsi défini, la diversité des communautés est un indicateur de la dégradation locale des cours d'eau.

Le quatrième chapitre consiste en la mise au point (jeu de données de construction) et le test (jeu de données additionnel) d'un indice multimétrique de la qualité écologique des cours d'eau de Guyane. Notre objectif était d'assembler des métriques présentant une forte discrimination (écart référence-perturbé), une faible spécificité vis-à-vis des types de perturbation (indice généraliste), une faible redondance entre métriques (complémentarité des informations écologiques), et bien entendu une faible variabilité en conditions de référence. Dans le cadre du projet PME soutenu par la DEAL Guyane, l'Office de l'Eau et l'ONEMA, les connaissances fondamentales (schémas typologiques des chapitres II et III) et l'outil (chapitre IV) rapportés dans cette thèse ont d'ores et déjà conduit à un transfert vers le domaine opérationnel via des programmes de mise en œuvre de la DCE dans les DOMs. En particulier, l'Indice Biologique Multimétrique Guyane (IBMG) va entrer dans le processus de validation par les partenaires susnommés puis de mise en œuvre.

Le cinquième chapitre nous a permis de nous concentrer sur un ordre spécifique d'insecte aquatique (Ephéméroptères) dans l'optique d'une évaluation de la qualité ciblée sur une pollution typique de la Guyane : l'orpaillage. Bien que critiquable par certains aspects et inadapté aux exigences de la DCE, l'indice SMEG développé par Alain Thomas (2001) avait révélé la pertinence des Ephéméroptères en tant que bioindicateurs de la qualité des cours d'eau de Guyane. Nous avons souhaité approfondir cette piste par l'utilisation de traits bioécologiques des 35 genres rencontrés sur les PME. Nous avons prédit que les perturbations génèrent des changements dans la combinaison de traits / fonctions écologiques représentés par les assemblages d'éphéméroptères à mesure que certaines espèces sont éliminées ou remplacées par d'autres. Ce travail a également été l'occasion de quantifier, pour la première fois, un ensemble de traits biologiques susceptibles de répondre à la dégradation des conditions physico-chimiques en Guyane. Les résultats, qui montrent une réponse des traits de vie non seulement aux perturbations en cours mais aussi passées (orpaillage ancien), encouragent la poursuite de travaux fondamentaux sur la biologie des invertébrés d'eau douce des petits cours d'eau de Guyane.

Le sixième article présente l'Indice Biologique Macroinvertébrés de Guyane et discute ces limites. Les DOMs représentent un enjeu majeur pour l'application de la DCE en France. Calibrée au départ pour des conditions tempérées, cette loi européenne pose de nouveaux défis scientifiques pour son application en milieu tropical. Ce chapitre pourra faire office de guide méthodologique pour la transmission de l'outil biologique au domaine opérationnel.

Chapitre I. Les spécificités de la Guyane Française

Ce chapitre dresse une présentation générale de la Guyane Française et ses milieux aquatiques.

Les protocoles expérimentaux et les méthodes d'analyse de données plus spécifiques à certaines parties de ce travail de thèse seront décrits dans les chapitres correspondants.

I.1. LA GUYANE FRANCAISE

I.1.1. Géologie et climat

La Guyane s'étend entre 2 et 6° de latitude nord et entre 52 et 54° de longitude ouest, dans la zone équatoriale de l'hémisphère nord, sur le continent sud-américain. Elle appartient à un ensemble géologique vaste, nommé « Plateau des Guyanes » ou « Bouclier guyanais » qui correspond à une formation du précambrien, d'âge compris entre 2,5 et 1,9 milliard d'années (Choubert, 1974; Milési et al., 1995). Le territoire Guyanais couvre approximativement 84000 km², borde l'Océan Atlantique sur 370 km et compte 22 communes (Figure 6). Il est délimité par 2 fleuves frontaliers : le fleuve Maroni, de 520 km de longueur, symbolise la frontière avec le Surinam et le fleuve Oyapock (403 km) qui symbolise la frontière avec le Brésil, avant de se prolonger au sud. Dans l'ensemble le relief de Guyane est très peu marqué, avec une orientation générale sud - nord, sauf pour quelques chaînons qui culminent à 800m (Figure 6), d'où une pente généralement faible des cours d'eau et une influence forte de la marée sur les zones côtières. Les formations du substratum assez récentes (Paléoprotérozoïque (2 à 2.2 milliards d'années) subissent encore une très forte altération (transformations physique et chimique de la roche initiale liées aux importantes conditions hydrolysantes du climat neotropical) et sont recouverts par des formations latéritiques de plusieurs mètres à plusieurs dizaines de mètres d'épaisseur (Nontanovanh et Roig, 2010). Ces sols latéritiques sont des sols pauvres en silice et en éléments nutritifs fertilisants (Ca, Mg, K, Na). Si l'on fait abstraction des alluvions récentes et actuelles que l'on peut observer le long des fleuves, les formations sédimentaires de Guyane sont strictement confinées sur la frange côtière où elles représentent une bande de cinq kilomètres jusqu'à une cinquantaine de kilomètres de largeur. L'épaisseur totale de ces sédiments est comprise entre quelques mètres et une cinquantaine de mètres localement. En résumé, les roches très anciennes du Bouclier guyanais, de type siliceuse, lessivées depuis 2 milliard d'années, fournissent très peu d'ions. Même les sables côtiers issus des latérites sont très peu solubles (sable siliceux).

Le climat en Guyane est de type équatorial humide. La position proche de l'équateur, ainsi que sa façade océanique, confèrent une bonne stabilité climatique avec des températures assez constantes de 25 à 26°C. Des minima de température sur l'intérieur du pays en novembre et décembre et des maxima en période sèche sont toutefois enregistrés. La moyenne mensuelle des minima et maxima sont compris entre 20,3°C (19,7-21°C) et 33,5°C (32,1-33,5°C) .

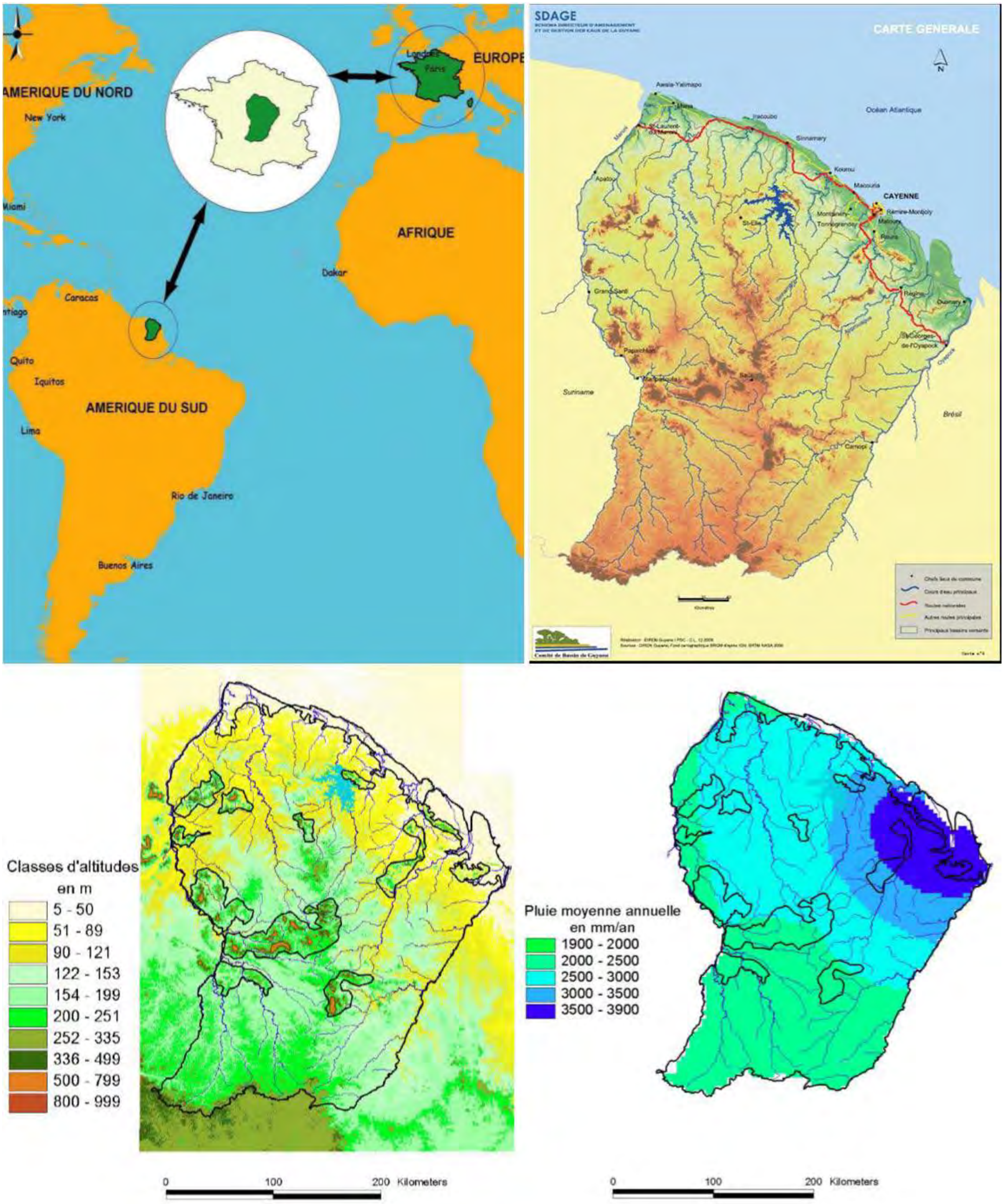


Figure 6: En haut : Localisation de la Guyane Française et organisation du territoire. En bas ; Relief et pluviométrie de la Guyane Française (source : Chandesris et al. 2005).

En général, nous observons une grande régularité des vents et des températures, qui varient faiblement au cours de l'année. Seules les précipitations connaissent des variations annuelles conséquentes, et c'est donc principalement ce paramètre météorologique qui détermine le rythme des saisons guyanaises.

On distingue l'alternance de quatre saisons en Guyane :

- La petite saison des pluies : vers la mi-décembre. Les pluies sont abondantes et la couverture nuageuse quasi permanente. Durant cette saison, l'amplitude des températures est faible, conséquence d'une insolation minimale.
- Le petit été de mars : Cette petite saison sèche constitue une trêve des précipitations. Cette période se traduit par un ciel en général ensoleillé et quelques averses localisées.
- La saison des pluies : Dès la fin du mois de mars, les précipitations redeviennent fréquentes. Ces périodes alternent avec des accalmies de quelques jours. Dans ces conditions, le temps devient variable et les éclaircies alternent avec des averses brèves mais souvent intenses.
- La saison sèche : La période vraiment sèche s'établit de la mi-août à fin octobre. Les journées sont bien ensoleillées avec quelques averses éparses.

La pluviométrie est cependant variable sur le territoire et se situe entre 2000 mm/an (sud et ouest) et 4000 mm/an sur une zone plus arrosée au nord-est (Figure 6).

I.1.2. Le réseau hydrographique

Du fait de sa position dans la zone climatique équatoriale humide, le réseau hydrographique guyanais est très dense. La mise en œuvre du référentiel BD Carthage® en 2013 (Mourguiart et Linares, 2013) a permis d'homogénéiser la connaissance géographique du réseau à l'échelle de la Guyane. Ce référentiel répertorie 112 000 km de cours d'eau et distingue en Guyane quatre régions hydrographiques majeures : le bassin versant du Maroni, le bassin versant de la Mana, les fleuves côtiers (Sinnamary, Comté, Kourou, Orapu, Tonegrande, Korossibo), le bassin versant de l'Approuague et le bassin versant de l'Oyapock (Figure 7). L'ensemble des fleuves de Guyane se jette au nord du district, dans l'océan Atlantique. Leurs débits présentent des variations annuelles quasi uni-modales avec des hautes eaux en mai et un étiage marqué en octobre (Figure 7). Cette tendance annuelle est toutefois marquée par une légère baisse des débits durant la période dite du « petit été de mars ». Les grands cours d'eau guyanais sont jalonnés de nombreux sauts, plus ou moins prononcés selon la saison, alternant avec des tronçons d'eau plus calme.

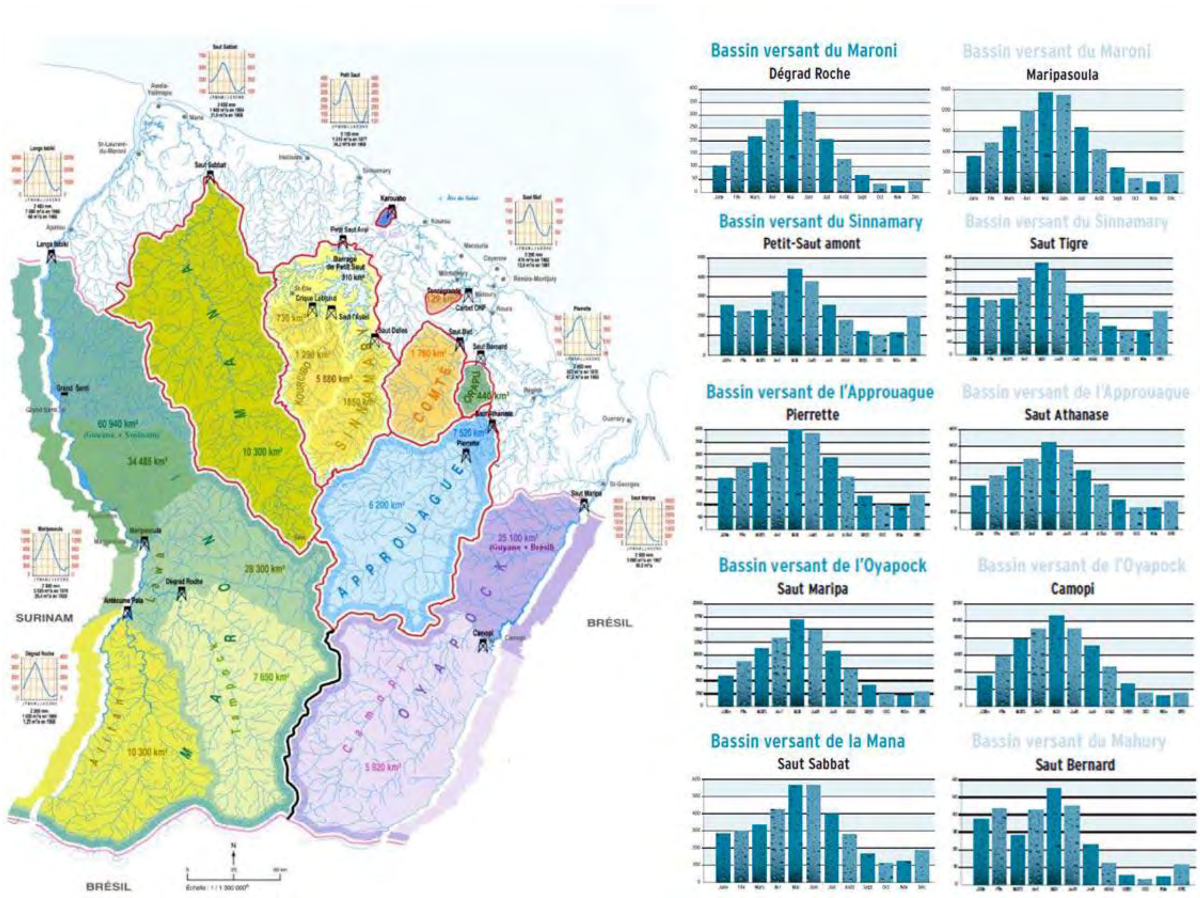


Figure 7 : Les principaux bassins de la Guyane Française (Atlas de Guyane, 2003) et débits moyens mensuels (période d'observation : 8 à 39 ans selon les bassins Monfort et Ruf, 2005).

I.1.3. Les petites masses d'eau (PME)

Les petites masses d'eau (PME) sont les cours d'eau de faible profondeur (rarement supérieure à 1m) et de faible largeur (inférieure à 10m). Ce type de masses d'eau représente **plus de 80% du linéaire total** du réseau hydrographique guyanais et se rencontre sur l'ensemble du territoire (Figure 8). Les connaissances actuelles des PME sont partielles (hydro-géomorphologie, fonctionnement écologique…). Ces masses d'eau ont été définies en utilisant les rangs de Strahler (indication de la taille du cours d'eau et sa place dans le réseau) à partir de la seule source disponible pour le réseau hydrographique, à savoir une digitalisation de la carte IGN au 1/500.000ème (« rang 500 »). Pour base de comparaison, la base cartographique utilisée en métropole pour définir les rangs est celle de la BD Carthage au 1/50.000ème. Il y a donc une forte différence de taille pour un même rang entre la Guyane et la métropole. De nombreuses PME de tête de bassin sont donc invisibles pour les images aériennes ou satellitaires du fait de la couverture forestière. La seule base pour améliorer la connaissance du réseau hydrographique serait la constitution d'un réseau théorique à partir d'un modèle numérique de terrain (RHT – Pella et al., 2012) qui pourrait ainsi définir affecter en fonction de l'écoulement une surface théorique minimum pour définir le début d'un réseau hydrologique (cours d'eau d'ordre 1). Actuellement, on ne dispose en Guyane que de modèles numériques d'élévation (MNE), qui correspondent à la surface de la canopée. A titre d'exemple, plus de la moitié des sites échantillonnés au cours de cette étude ont été fait sur des cours d'eau non répertoriées.

Figure 8 : Les petites masses d'eau : Localisation sur le réseau hydrographique (DEAL, 2014) et morphologie d'un tronçon (Crédit photo : N.Dedieu)

La chimie des eaux guyanaises est majoritairement contrôlée par la géologie. La conductivité est extrêmement faible (dans la gamme 20-80 µS.cm-1 avec des maximums dans le centre); et même la remise en suspension des sédiments (orpaillage) ne modifie que très faiblement la conductivité. Les petites masses d'eaux sont naturellement très peu chargées en particule (MES < 7 mg/L) et très peu turbide (< 3 NTU). Ces caractéristiques sont également retrouvées dans les petits cours d'eaux du bouclier Guyanais (Hammond et al., 2007). Les nutriments (phosphate et Nitrate) sont en limite de détection dans les milieux naturels (De Merona et al., 2001) et très peu présent même dans les zones urbanisées (maximum mesuré : 0.83 µg de NO3/L). Les eaux sont par endroit très acides : le pH mesuré dans les petits cours d'eaux de Guyane est compris entre 4,04 et 7,6. Les eaux les plus acides correspondent aux « Eaux noires » dans la typologie Amazonienne (« Amazonian black water » – Sioli, 1967, 1968). Cela est lié au couvert forestier qui apporte des matières organiques composées d'acides humiques non dégradables. Les petites masses d'eaux peuvent donc être caractérisées comme « ultraoligotrophes, acides » sur le point chimique et pourrait de ce fait être considérées comme peu favorables aux organismes aquatiques (« peu biogènes »).

En ce qui concerne l'hydromorphologie des petits cours d'eau, la présence de radiers est rare. Cela s'explique par le relief relativement plat, et par conséquent le courant est faible. Contrairement à l'Europe ou aux Antilles où la gamme de vitesses de courant est comprise entre 0 et 150 $cm.s^{-1}$ (Souchon et al., 2000 ; Bernadet et al., 2013) , le courant dans les petites masses d'eaux excède rarement 75 $cm.s^{-1}$ et est plus généralement compris entre 0 et 50 $cm.s^{-1}$. L'hydrologie est caractérisée par l'alternance saison humide (Décembre - Aout) et sèche (Septembre - Décembre). En conséquence, l'hydraulicité est très forte pendant la saison des pluies et très faible (et plus stable) pendant la saison sèche. La roche en place produit essentiellement des sables assez grossiers, mélangés sur les petites masses d'eau à des graviers en cours d'altération. On rencontre donc en général un substrat sableux, mobile, peu biogène dont les seuls éléments stables sont les bois immergés. Les minéraux grossiers (de type galet ou bloc) sont très rares en Guyane. Les macrophytes ont été peu rencontrées dans les petites masses d'eaux ; cela est surement lié à ces substrats sablo-vaseux très mobiles et la faiblesse naturelle des intrants nutritifs azotés et phosphorés. Dans certaines PME boisées, le lit du cours d'eau peut être entièrement recouvert de litières en faisant l'habitat le plus biodisponible au niveau du site. Les racines sont considérées comme un habitat abondant par rapport aux autres types et peuvent être observés sous deux formes : tapis racinaires (au niveau des berges) et sous forme de « chevelus » (racines tombantes (ex : liane)). Les chevelus sont considérés comme un habitat très biogène.

Au cours des deux campagnes d'échantillonnage, trois faciès de ripisylve ont été observés : les forêts de *terra firme,* les *forêts de bas-fond* et les *forêts de sable blanc* (Figure 9). Ces ripisylves présentent différentes structures végétales due aux conditions environnementales contrastées en termes de ressources en nutriments, de contraintes édaphiques et climatiques (Granville, 2002). Les socles de sable blanc sont généralement couplés aux rivières d'eaux noires, parce que la végétation qui pousse sur ces sols est exceptionnellement riche en acides humiques (= tanins et autres composés phénoliques).

Figure 9 : Les trois types de ripisylves rencontrées au niveau des petites masses d'eau : (a) les forêts de terra firme, (b) les forêts de bas-fond ou inondées et (c) les forêts de sable blanc.

I.1.4. Typologie des PME

La typologie (ou classification) permet de classer des masses d'eaux en groupes homogènes du point de vue de certaines caractéristiques naturelles. Elle est l'élément essentiel permettant de définir les conditions de référence et le bon état écologique, qui sont établis par « type de milieu » (regroupement de sites présentant des caractéristiques communes). ***Cette classification « a priori*** » est établie à partir des connaissances et d'hypothèses sur les facteurs de contrôle de la biodiversité des communauté (les facteurs de contrôle étant d'une manière générale l'habitat physique à l'échelle locale, et à plus large échelle l'hydrologie, la géomorphologie des cours d'eau, la végétation riveraine, la géologie, le relief et le climat) (Figure 10).

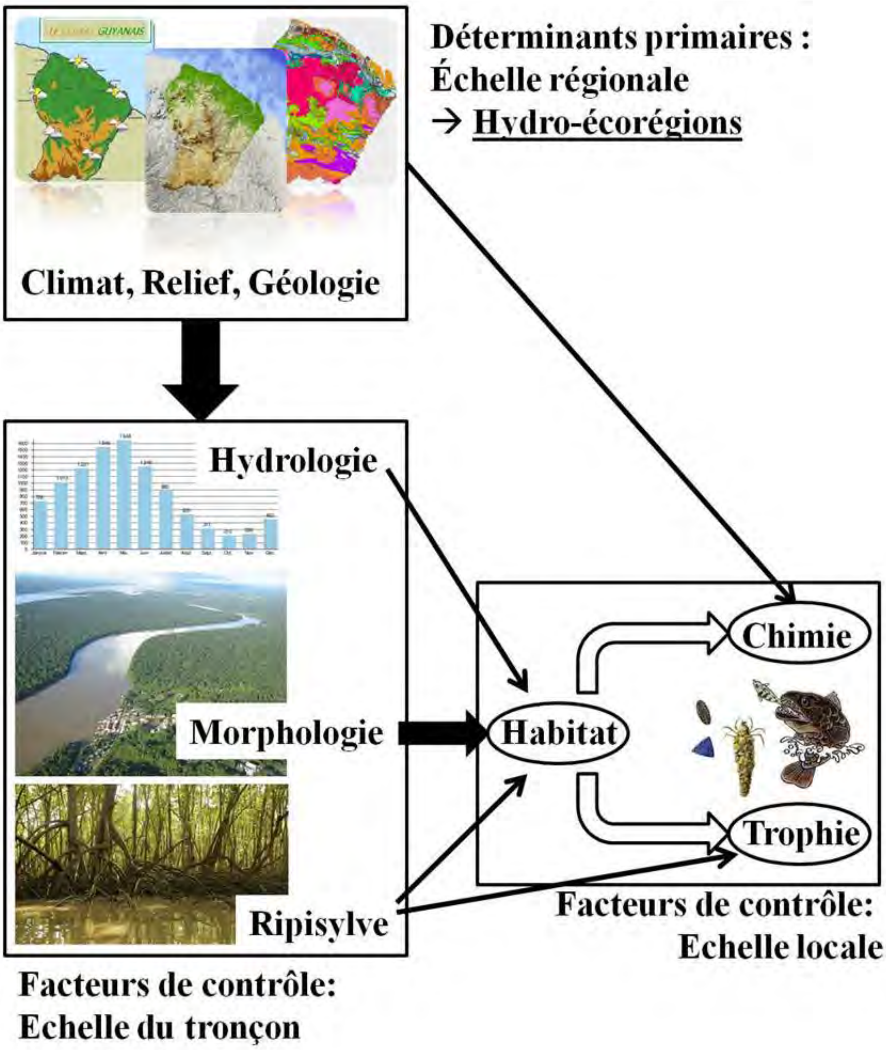


Figure 10 : Les Hydro-écorégions. Représentation schématique de l'emboitement hiérarchique des facteurs de contrôle des écosystèmes d'eau courante. (Schéma modifié de Chandesris et al., 2005).

Une première classification *a priori* a été établie à l'échelle nationale (métropole et DOM) par le Cemagref en 2005 (Chandesris et al., 2005). Cette typologie des masses d'eaux, appuyée sur la distinction géologique et dans une seconde mesure sur le relief, a définie deux grandes hydro-écorégions : le « **bouclier guyanais** » et « **la bande côtière** » (Figure 11). Cette typologie a été affinée par des classes de taille de cours d'eau définies comme en métropole (rangs Strahler).

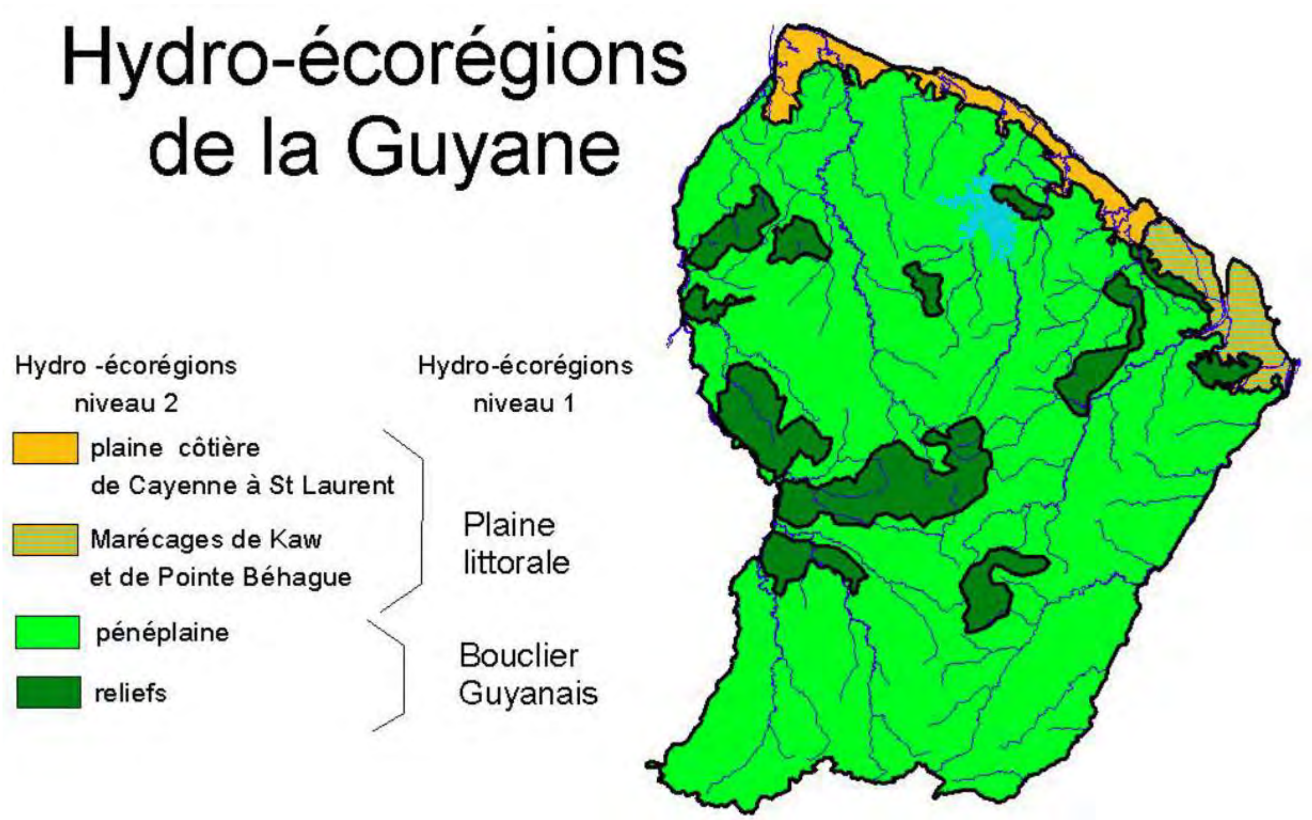


Figure 11 : Les Hydro-écorégions de Guyane (Chandesris et al., 2005).

La typologie « *a priori* » peut ensuite être validée par une **biotypologie** (Bernadet et al., 2013). Ce type de ***classification « a posteriori »*** est obtenu à partir du traitement statistique de données sur la distribution de variables biotiques et/ou abiotiques. Cette organisation permet d'avoir une image bien représentative des peuplements de références attendues par **types de milieu**. Elle permet aussi de tenir compte de la variabilité environnementale des communautés et de s'en affranchir dans une certaine mesure. Les données physico-chimiques, géomorphologiques et les données relatives à l'occupation du sol ont été associées aux données biologiques afin de dégager les profils de distribution des macroinvertébrés en fonction des conditions environnementales. Au début de cette étude, nous ignorions l'importance de la géomorphologie sur la distribution des invertébrés, et dans quelle mesure les perturbations anthropiques surpassent les facteurs géomorphologiques dans la distribution des macroinvertébrés à l'échelle de la Guyane. Afin de déterminer l'importance relative des variables géomorphologiques et des perturbations anthropiques sur la distribution des espèces, des comparaisons ont été réalisée. Cette typologie a fait l'objet d'une publication (Chapitre III).

I.1.5. Les invertébrés benthiques de Guyane Française

Des travaux antérieurs sur le territoire (De Merona, 2001 ; Thomas et al., 2001 ; Wasson et al., 2008), nous ont permis de dresser un premier bilan des caractéristiques locales qui sont en prendre en compte pour l'étude de la macrofaune benthique guyanaise:

- La très faible densité de la macrofaune benthique, même dans les milieux de référence, avec quelques centaines d'individus par m^2, dont 50 à 80 % de Chironomidae ; et des organismes de très petite taille ;
- Une diversité relativement faible au niveau d'une station, de l'ordre de 20 à 25 familles dans les sites de référence;
- Très peu d'habitats colonisés. Les dépôts de feuilles mortes, les bois immergés abritent très peu d'individus. Les sables et les roches sont pratiquement vides de faune.
- Il y a une quasi-absence de déchiqueteurs ;
- Il y a seulement une espèce de Plécoptère (*Acroneuria pectipes*).

Ces caractéristiques faunistiques peuvent s'expliquer par le caractère ultra-oligotrophe des eaux guyanaises (voir ci-dessus). Il est possible également que dans des milieux aussi pauvres, l'effet de compétition et de prédation par les poissons soit très important (Blumenshine et Kemp, 2000). Il faut signaler également que l'on présente ici des observations principalement faite au niveau taxonomique de la famille et que la diversité spécifique à l'échelle de la région reste encore à étudier. Les principales (et quasi-uniques) connaissances sur la taxonomie de la faune invertébrés proviennent du projet « Qualité des Eaux de Rivières de Guyane » effectué en 1999 (De Merona et al., 2001). Le but de ce projet était de mettre en place des inventaires préliminaires dans le but de créer un outil biologique pour les rivières de Guyane. Ces travaux ont démontré que deux ordres étaient surreprésentés dans les rivières de Guyane Française: les Diptères et les Ephéméroptères. C'est ce dernier ordre qui avait été retenu pour élaborer le premier (et actuellement unique) indice de qualité pour les eaux guyanaises. Cet indice, intitulé «SMEG » (Score Moyen des Ephémères de Guyane - Thomas *et al.*, 2001) est basé sur le concept des scores (ASPT : *Average Score Per Taxa*- Armitage et al., 1987), c'est à dire sur l'addition des coefficients de polluo-sensibilité attribués aux différents genres. A chaque genre est attribué une note de polluo-sensibilité (exemple : 1 = peu à 5 = très polluo-sensible). Cependant, cet indice préliminaire souffre d'un défaut de conceptuel de construction qui consiste à considérer les espèces vivant en tête de bassin comme « polluo-sensibles » et celles qui vivent dans les zones aval comme « résistantes » (Tableau I). De ce fait, même dans des milieux non impactés, la valeur brute de l'indice diminue de l'amont vers l'aval.

Tableau I : Barème d'évaluation du Score Moyen des Ephéméroptères Guyanais (Thomas, 2001)

SMEG	Classe	Etat des cours d'eau
i > 4.5	I	Rivières de faible largeur ou petites rivières sans impact anthropique notable.
4 > i > 4.5	II	Rivières faiblement impactées, ou stations suffisamment éloignées des impacts.
3 > i > 4	III	Influences anthropiques durables mais d'intensité moyenne.
2 > i > 3	IV	Fleuves et larges rivières exposées à des impacts anthropiques aigus.
1 > i > 2	V	Pollutions importantes; fort déficit en 0_2 et/ou substratum très modifié.
0	VI	Cours d'eau dépourvus de macroinvertébrés polluo-sensibles (EPT).

De plus, l'évaluation actuelle du statut écologique par cet indice ne peut être considérée totalement DCE-compatible parce que l'information prise en compte n'est pas quantitative et que le jugement qualitatif n'est pas exprimé par rapport à un état de référence (EQR).

I.1.6. Les Ephéméroptères de Guyane Française

Comparée aux autres groupes systématique du benthos d'eau douce, la connaissance taxonomique des Ephéméroptères de Guyane Française a grandement progressé au cours des dernières années (avec 4 genres signalisés en 1998, 37 nommées en 2001 (Thomas et al., 2001 - Figure 12). Néanmoins, l'observation de nouveaux genres au cours de notre étude (*Tricorythopsis, Euthyplocia* (à confirmer)) démontre qu'il reste toutefois des progrès à accomplir dans l'inventaire des taxons guyanais et particulièrement au niveau des petits cours d'eau. En effet, l'état d'avancement des inventaires actuels montre que près de 1% des 442 espèces d'éphéméroptères connues en Amérique du Sud sont rencontrées en Guyane Française, par rapport à 38% et à 30% au Brésil et en l'Argentine, respectivement. Les petits pays de la région tels que le Pérou ou l'Equateur abritent deux fois plus d'espèces d'éphéméroptères que la Guyane Française ou encore le Venezuela (Chacón et al., 2009). Domínguez et al., (2006) suggèrent que ces énormes écarts de composition faunistique découlent probablement plus de l'histoire de collecte plutôt que la réelle richesse taxonomique (Pescador et al., 2001).

SCORE individuel (provisoire)	43 UNITES OPERATIONNELLES (dont 42 Genres)	
1	**Très peu polluosensibles (4 U. O.)**	
	Caenis	*Callibaetis*
	Aturbina	*Cloeodes*
2	**Peu polluosensibles (8 U. O.)**	
	Asthenopus	*Lentvaaria*
	Campsurus	*Apobaetis*
	Bessierus	*Harpagobaetis*
	Hydrosmilodon	*Paracloeodes*
3	**Assez polluosensibles (10 U. O.)**	
	Hexagenia	*Americabaetis*
	Tricorythodes	*Camelobaetidius* à paracerque long
	Hermanella sensu lato	*Spiritiops*
	Simothraulopsis	*Waltzoyphius*
	Terpides	Oligoneuriidae *gen. sp.* 2
4	**Polluosensibles (9 U. O.)**	
	Brachycercus	*Microphlebia*
	Leptohyphes	*Camelobaetidius* à paracerque court
	Fittkaulus	*Tomedontus*
	Hagenulopsis	*Zelusia*
		Oligoneuriidae *gen. sp.* 1
5	**Très polluosensibles (12 U. O.)**	
	Campylocia	*Genre X*
	Coryphorus	*Genre Y*
	Miroculis	*Adebrotus*
	Thraulodes	*Cryptonympha*
	Ulmeritoides	*Guajirolus*
	Genre U	*Rivudiva*

Figure 12 : Ephéméroptères de Guyane et leurs valeurs indicatrices (Thomas, 2001)

La déforestation et l'orpaillage ont été démontrés comme les principales menaces sur les communautés d'Ephéméroptères au niveau des tropiques (Benstead et al., 2003; Benstead et Pringle, 2004 ; Dudgeon, 2000). Ainsi, une étude spécifique cette ordre a été menée afin d'évaluer la capacité indicatrice des éphéméroptères en Guyane Française (Chapitre V). Pour cela, des informations sur leur biologie ont été rassemblées par des recherches bibliographiques et des analyses en laboratoire. Codées sous forme de traits biologiques, ces informations biologiques permettent d'analyser le potentiel des espèces en matière de croissance, de mode de nutrition (ex. taille, stockage), de reproduction (ex. soins aux jeunes), de mobilité (ex. fuite, migration, diapause) et plus généralement en terme de capacité de résistance ou de résilience aux perturbations naturelles ou anthropiques (Charvet et al., 1998 ; Doledec, 2009). Dans notre étude, 5 traits biologiques, divisés en 21 modalités ont été décrits. Le codage utilisé est appelé codage « flou » : pour chaque modalité de trait, un score de 0 à 3 est attribué au taxon suivant le degré d'affinité du taxon pour la modalité. Ce type de codage permet de tenir compte de la variabilité biologique des invertébrés qui ne pourrait être complètement retranscrite au travers d'un simple codage binaire (Chevenet et al., 1994). Les traits sélectionnés sont :

- La Taille maximale potentielle: La taille maximale potentielle correspond à la taille la plus grande atteinte par les organismes au cours de leurs phases aquatiques. Les tailles des éphéméroptères rencontrées étaient généralement comprises entre 0.25 et 1.5 cm.
- La Forme du corps : Ce trait biologique a été décomposé en 3 modalités : corps plat, hydrodynamique ou cylindrique. Les différentes modalités peuvent nous renseigner sur une potentielle adaptation aux contraires hydrologiques (ex : les corps plat ou hydrodynamique sont généralement plus adaptés au fort courant),
- Les formes de branchies : Les branchies ont été classées en 5 modalités (Figure 13). Le type de branchie renseigne sur les capacités respiratoires des individus (ex : capacité d'assimilation de l'oxygène, protection face à une augmentation des matières fines qui pourraient obstruer les branchies). Ce trait peut être intéressant parce qu'il peut expliquer la disparition de certains taxons dans des milieux pauvres en oxygène,

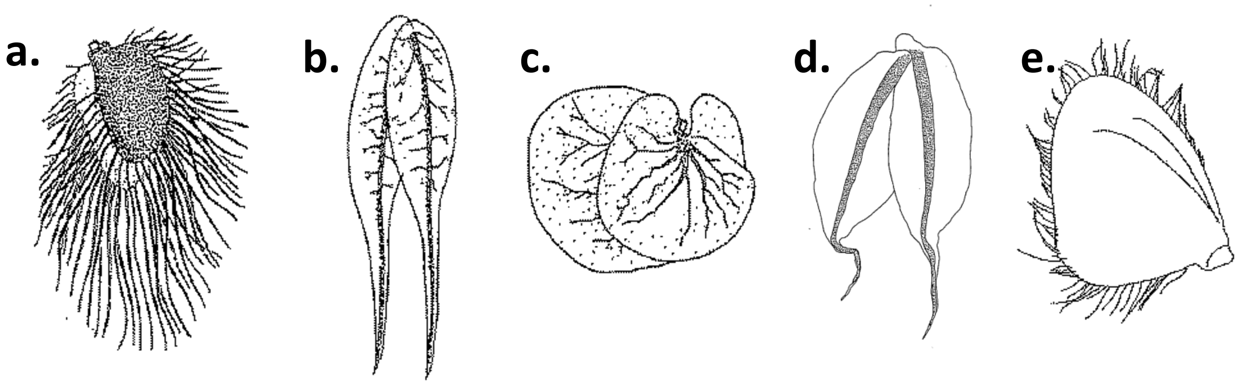


Figure 13: Les différents types de branchies observées entre les différents genres d'Ephéméroptères.

- Le groupe trophique regroupe 5 modalités : Broyeurs (Shredder), collecteurs (Coga), filtreurs (CoFi), herbivores (Scraper/Brusher). Ces informations nous renseignent sur les exigences alimentaires des individus et par conséquent sur le lien avec les ressources disponibles dans le milieu. Le codage flou s'applique bien à la détermination de la trophie car une description d'un trait d'une espèce par un système binaire aboutirait à la sélection d'une unique source de nourriture pour ce taxon, et conduirait donc à une large imprécision quant à la description du régime alimentaire des espèces les plus omnivores.
- Type de locomotion : Ce trait a été découpé en 4 modalités : nageur, rampeur, fouisseur (substrat de surface ou profonde). Ce trait nous renseigne sur la capacité d'un individu à se déplacer dans son milieu. La modification de l'hydromorphologie du cours d'eau peut agir comme un filtre de sélection et entrainer la disparition des individus aux capacités de locomotion les moins adaptées.

I.2. LES PRESSIONS HUMAINES SUR LES MILIEUX AQUATIQUES

I.2.1. La population Guyanaise

Avec 90.000 km^2 et environ 250.000 habitants, la Guyane est le plus grand département de France, mais aussi le moins peuplé. La densité de population est de 2,1 habitants/km^2, dont 60% à Cayenne et 80% dans les trois plus grandes agglomérations (Figure 14 : Cayenne, Kourou et St Laurent du Maroni). Cependant, la Guyane présente un taux de croissance moyen depuis 1999 de 3,7 % par an, soit deux fois plus que la Réunion et cinq fois plus que l'hexagone. La Guyane, qui comptait 229 040 habitants en 2010 (Prud'homme et Treyens, 2010) pourrait en compter plus de 700 000 en 2040 (Horatius-Clovis, 2011). Pour pallier à cette expansion démographique, les besoins estimés de production de logements s'élèvent à environ 4400 logements/an d'ici 2040 (Yahou-Dauvier et Planchat, 2014). Les villes de l'intérieur du pays sont autant concernées par cette expansion démographique que celles du littoral, la population prévue de Maripasoula située sur le haut Maroni serait de 71.000 habitants en 2030, soit 54.000 de plus qu'en 2013, ce qui entrainera probablement un impact environnemental non négligeable dans ces zones encore peu peuplées. A la population dénombrée par l'Insee, il faut ajouter une population clandestine non négligeable, provenant des pays frontaliers à la Guyane (Brésil, Surinam). En décembre 2010, le nombre de clandestins en Guyane était estimé entre 30.000 et 60.000.

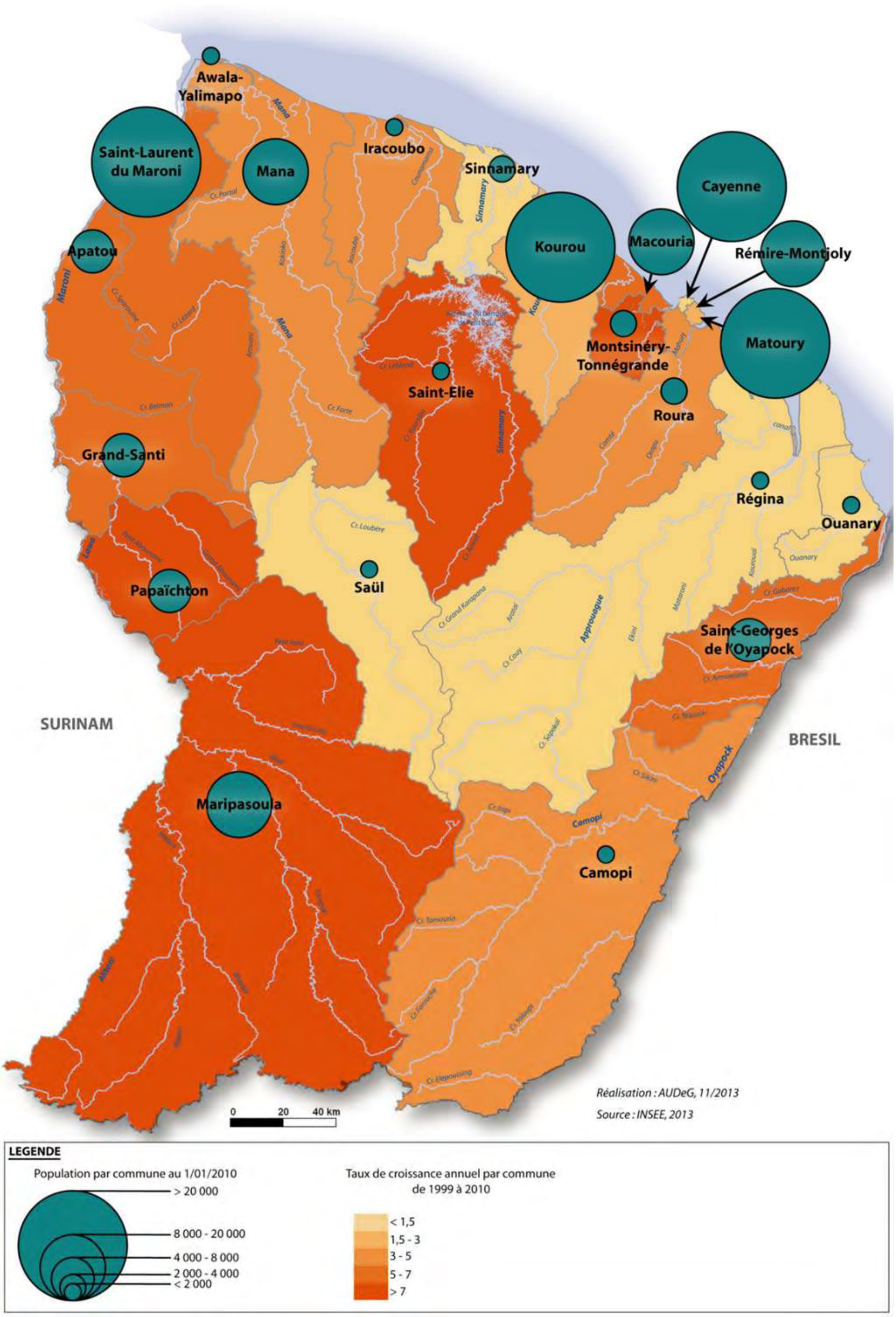


Figure 14 : La démographique de la Guyane Française (SAR, 2014)

I.2.2. La bande côtière

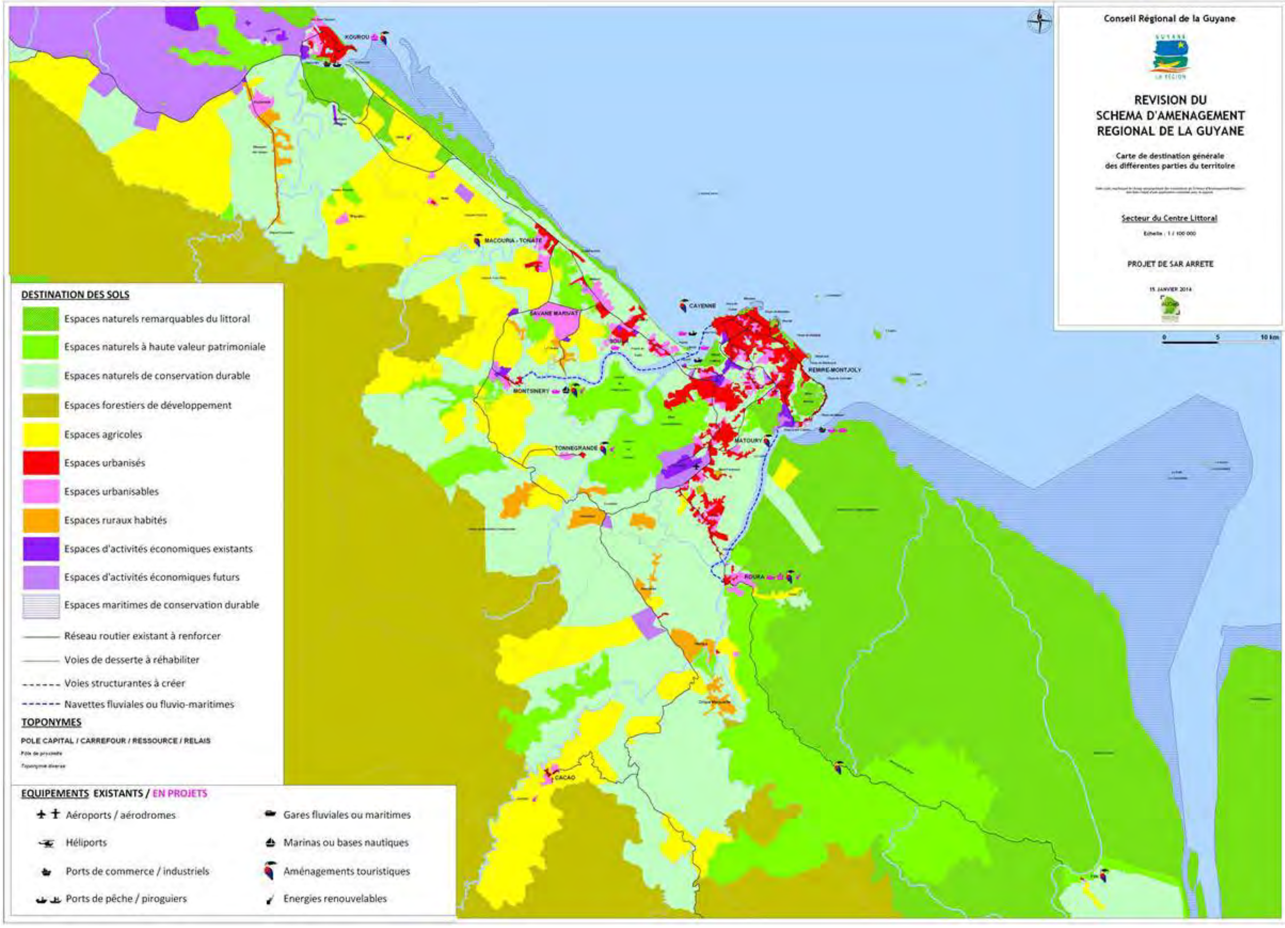


Figure 15 : La bande littorale guyanaise - Occupation des sols

La bande côtière est actuellement le secteur de développement prioritaire du territoire guyanais. Elle ne représente que 8% du territoire, mais concentre 80% de la population dont 103000 habitants sur l'île de Cayenne et 51000 autour de Saint Laurent, sur le Maroni, sur les 250000 habitants que compte actuellement la Guyane (Figure 14 et 15). Deux routes principales longent la côte sur 440 km et connectent les 15 communes littorales. Celles-ci concentrent les structures urbaines et industrielles principales. Un réseau routier secondaire permet d'accéder aux espaces moins urbanisés de la plaine littorale, composés de zones d'exploitation agricole et forestière, ainsi que de savanes, marais et mangroves (Figure 15). Ce réseau secondaire se compose de 530 km de voiries communales, 100 km de pistes agricoles et 1260 km de pistes forestières (accessibles uniquement à l'aide de véhicules spécialisés de type 4x4), éloignées de 70 km au maximum de l'océan, ce qui limite l'accès routier à la bande côtière (Geffrin et Labia, 2011). La faible couverture géographique du réseau routier explique en partie l'état de conservation exceptionnel de la forêt primaire de l'intérieur de la Guyane. Les activités humaines principalement concentrées sur la bande littorale sont trois sources de pression bien identifiées (Figure 15):

- la pollution urbaine, incluant une faible activité industrielle ;
- les eaux domestiques issues d'habitations éparses, hors agglomérations, qui se rejettent sans traitement dans des estuaires ou dans des cours d'eau littoraux ;
- les activités agricoles.

I.2.2.1. Activités agricoles

Les espaces agricoles ne recouvrent que 23 176 ha, soit 0,3 % du territoire guyanais (ONF, 2013). L'agriculture traditionnelle et familiale sur « abattis » représente les 3/4 des exploitations agricoles de Guyane *(*Agreste, 2000 ; DAF Guyane*)*. L'abattis traditionnel se traduit par une alternance de cycles de plantation et de jachères. Le cycle d'un abattis commence avec l'abattage des gros arbres et du sous-bois en début de saison sèche, soit en juillet-août. Ensuite vers octobre-novembre, l'agriculteur va procéder au brûlis. A l'aide d'une torche et parfois même d'un bidon d'essence, il va pouvoir brûler un à deux hectares par jour. Les parcelles d'abattis vont rarement dépasser les trois hectares. Ce système de culture sur brûlis est pratiqué par tradition pour déforester (troncs, sous-bois et herbes) la parcelle que l'on va cultiver, mais aussi pour fertiliser le sol avec le charbon de bois et les cendres produites, et pour tuer la faune du sol (Figure 16). Les abattis se développent en particulier au niveau des fleuves frontaliers et des alentours des villages amérindiens et bushinengues de l'intérieur ou du littoral. Le reste des activités agricoles se concentre principalement autour de 2 communes (Cacao et Javouhey) et alimente les marchés des villes côtières. Des rizières sont aussi présentes sur une partie de la plaine littorale, mais ne sont quasiment plus exploitées. Ces zones agricoles, de part leur faible couverture géographique, ont été peu rencontrées au cours de notre étude (n=4). Cependant, la Guyane connaît une augmentation de superficie de terrain agricole de 4,85 % par an, ce qui a représenté 8237 ha de surface agricole en plus entre 2005 et 2011. Cette activité est donc à prendre en compte car les impacts qui en découlent sur les milieux aquatiques sont multiples. Premièrement, le sol mis à nu est soumis à l'*érosion* directe du vent et des pluies et peut entrainer une augmentation de la turbidité. Ensuite, du fait du contexte agricole difficile (pauvreté du sol en nutriment, ravageurs, végétations indésirables), des intrants agricoles sont généralement utilisés (engrais, pesticide, fongicide, herbicide). Ces produits vont ensuite se retrouver dans les milieux aquatiques lors du lessivage des sols. L'impact écologique des produits phytosanitaires est reconnu mais difficile à évaluer du fait de la multiplicité et des interactions des produits et de leur large spectre d'action Enfin, pour irriguer les parcelles cultivées, des prélèvements d’eau sont généralement fait dans les petites masses d’eau et peuvent entrainer des assèchements pendant les périodes de basses-eaux.

Figure 16: Abattis traditionnels : parcelle pendant un brulis (en haut) et après un brulis (en bas) (Crédit photo : Dedieu, N.)

II.2.2.2. Activités industrielles

La Guyane possède un tissu industriel peu important. De nombreuses structures dites industrielles relèvent davantage de l'artisanat, et une grande partie des produits consommés sont importés de métropole. Les entreprises industrielles sont de petite taille sur le territoire : seulement 2 % des entreprises comprennent plus de 20 salariés, et 90 % ont moins de 6 salariés ((Prud'homme et Treyens, 2010). Les industries sont géographiquement concentrées sur l'île de Cayenne (66 % - Figure 17) et Kourou (16 %) et plus généralement dans les 15 villes côtières. Les perturbations engendrées par les pollutions des grandes agglomérations et activités industrielles ne sont pas prises en compte dans ce travail, car ces agglomérations se trouvent dans des zones ou les milieux aquatiques sont soumis à l'influence de la marée, et donc considérées comme des eaux de transition.

Figure 17 : Ville de Cayenne

I.2.3. Le plateau guyanais

A l'intérieur des terres, le contexte de la Guyane est singulier par rapport à la bande côtière mais aussi à la métropole ou aux autres DOM, du fait des ressources primaires abondantes dans les terres (bois précieux, or, bauxite) ainsi que des voies d'accès qui y sont peu développées. En effet, sept communes se sont développées à l'intérieur des terres et le long des fleuves (Figure 14). Ces communes ne sont pas accessibles par la route et sont seulement desservies par les voies fluviales et aériennes. L'intérieur des terres, dont la densité d'habitant est de l'ordre de 0,5 hab/km^2, appartient au domaine public de l'État et représente 80% de la superficie totale, dont la majeure partie dans le domaine public forestier. Les chantiers d'orpaillage et forestiers sont abondants à l'intérieur des terres et représentent donc les impacts majeurs sur les milieux naturels.

I.2.3.1. Pressions et impacts liés à l'exploitation forestière

I.2.3.1.1. L'exploitation forestière en Guyane Française

En moyenne sur les 10 dernières années, il a été produit 70000 m^3 de grumes par an (Berlioz, 2012). Ce chiffre semble actuellement à la hausse. Les exploitations sont actuellement localisées dans un rayon de 70 kilomètres de la côte, pour des raisons essentiellement économiques et logistiques (coûts d'exploitation, accès). L'activité se développe dans la zone correspondant au domaine forestier permanent, géré par l'ONF (Figure 18).

Depuis 2009, l'ensemble des exploitations forestières en Guyane doit intégrer des pratiques de gestion respectueuses de l'environnement énoncées par l'ONF dans « la charte d'exploitation à faible impact » (Panchout, 2010) qui impliquent des normes à respecter. Il s'agit d'une exploitation « extensive »; une trentaine d'espèces seulement (sur 1200) sont exploitées, à raison de 2 à 5 arbres à l'hectare. L'ONF conserve systématiquement, à proximité de chaque parcelle exploitée, des parcelles « de référence » intouchées. Une éco-certification est en cours de mise en place pour les exploitants. De plus, une distance minimale de 100 mètres du cours d'eau a été instaurée pour exploiter du bois. Les pistes créées doivent suivre le plus possible les lignes de crêtes pour ainsi éviter de traverser les rivières et créer des ouvrages de franchissement (Figure 19). Enfin, le travail d'extraction des arbres des parcelles ne peut être effectué qu'en période de saison sèche.

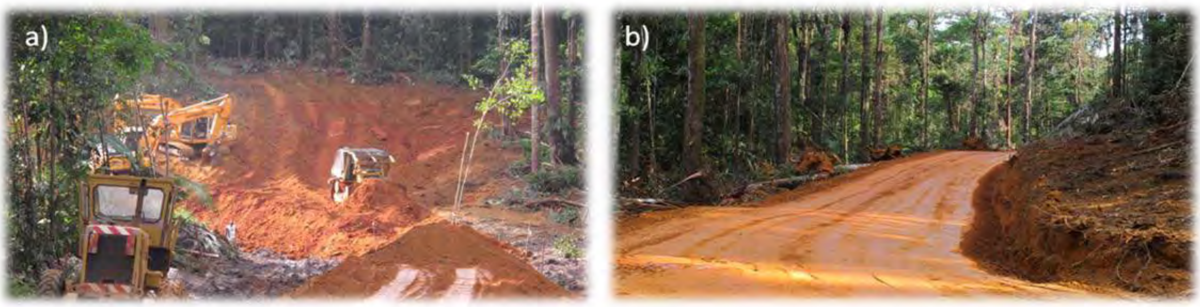


Figure 19: Aménagements engendrés par l'exploitation forestière :
a) création d'un pont pour traverser une rivière; b) piste forestière nouvellement crée.

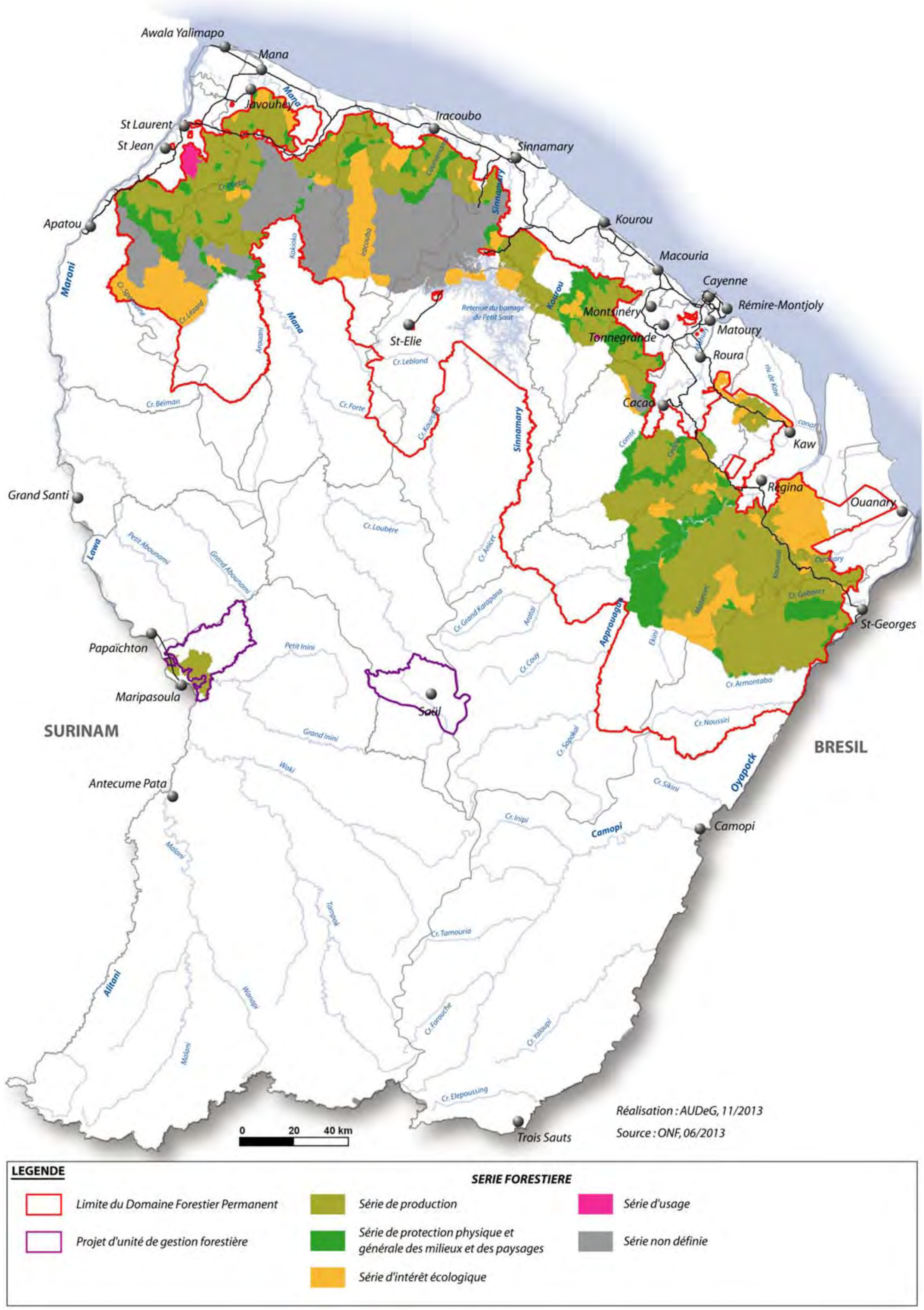


Figure 18 : Les zones d'exploitations forestières en Guyane (SAR, 2014)

*I.2.3.1.2***.** *Impact de l'exploitation forestière sur les milieux aquatiques*

L'impact de l'exploitation forestière a été bien étudié au cours des dernières années. La déforestation a pour conséquence de dégrader les habitats des cours d'eaux (Iwata et al., 2003; Wantzen, 2006), modifier le régime hydrologique et les ressources primaires (Benstead et al., 2003; Bojsen et Jacobsen, 2003; Benstead et Pringle, 2004) entrainant ainsi des changements majeurs de communauté benthique et une baisse de la diversité (Cummins et al., 1989 ; Sweeney, 1993 ; Benstead et al., 2003;. Bojsen et Jacobsen, 2003; Iwata et al., 2003; Dudgeon, 2006; Wantzen, 2006). Localement, les scieries peuvent aussi être une source de pollution du fait des traitements appliqués aux grumes et des rejets d'hydrocarbures dus à l'utilisation d'engins mécanisés. Des études récentes ont montré que la ripisylve est étroitement liée aux processus hydrologiques tropicaux (Heartsill-Scalley et Aide, 2003 ; McKergow et al., 2004; Gomi et al., 2006 ; Lorion et Kennedy, 2009). En effet, la déforestation a un impact majeur au niveau de l'interface terre/cours d'eau en réduisant les ressources allochtones, l'ombrage et en entrainant une sédimentation accrue (Gregory et al., 1991; Naiman et Décamps, 1997). Des travaux ont notamment démontrés qu'une zone riveraine boisée « tampon » réduit considérablement les impacts de la déforestation sur les cours d'eaux (Pringle et Scatena, 1999 ; Benstead et al., 2003 ; Lorion et Kennedy, 2009). C'est donc afin de limiter les impacts environnementaux que l'ONF a mis en place la charte d'exploitation à faible impact. Cependant, la déstructuration des sols lors de la création de pistes, du passage des grumiers ou des engins sur les pistes de débardage provoquent quand même une hausse de la quantité de sédiments, même si elle est limitée (Figure 19a et b). Cet apport sédimentaire supplémentaire reste un facteur perturbant important pour la faune aquatique, comme cela a pu être démontré lors d'une étude menée dans les rivières de la région de Manaus, au Brésil (Dias et al., 2010).

<u>I.2.3.2. Pressions et impacts liés aux activités aurifères</u>

I.2.3.2.1. L'or en Guyane Française

L'exploitation aurifère constitue le facteur de pression majeur en Guyane, et c'est même le seul facteur qui semble susceptible de provoquer une dégradation significative de l'état écologique. La pression d'orpaillage est fortement corrélée au cours de l'or. En effet, l'or ayant plus que sextuplé entre les années 2000 et 2012, l'orpaillage en Guyane est actuellement en plein essor, d'autant plus que les ressources aurifères inexploitées sont encore très importante (Figure 20). La superficie des formations géologiques favorables à la découverte de nouveaux gisements couvre 62 800 km², soit 75 % de la superficie de la Guyane (SDOM, 2011). L'or se présente sous forme de deux types de gisements :

- Les gisements primaires représentent les filons d'or natif contenus dans les roches situées à une profondeur comprise entre 20 et 100 mètres. L'exploitation de ces gisements est rare, car elle demande la création d'une mine à ciel ouvert à l'aide de moyens mécaniques importants, lourds et coûteux, qu'il faut acheminer en pleine forêt.
- Les gisements secondaires proviennent de la destruction progressive des gisements primaires par érosion. Les minéralisations aurifères des roches altérées se trouvent alors libérées sous forme d'éluvions (l'or reste à flanc de colline) ou d'alluvions (l'or libéré est entraîné par les eaux courantes et se retrouve dans le lit mineur et majeur des cours d'eau).

Figure 21 : Impacts et pressions de l'exploitation aurifère sur les cours d'eau guyanais : a) baranques. b) exploitation légale. C) exploitation illégale. d) rejet des boues dans le milieu naturel.

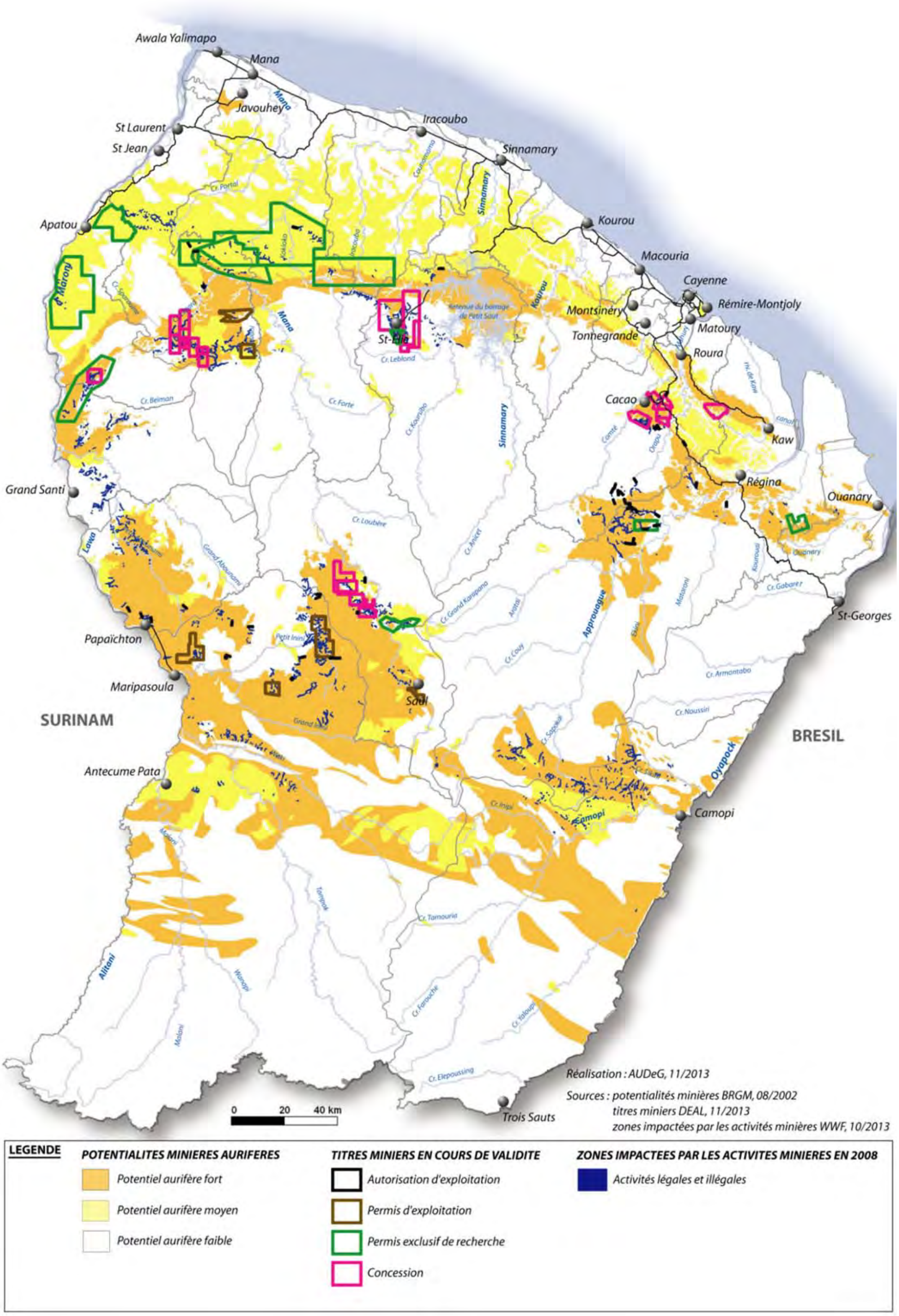


Figure 20 : Les potentialités minières aurifères en Guyane Française (SAR, 2014)

I.2.3.2.2. L'exploitation aurifère en Guyane Française

Les gisements alluvionnaires sont les plus exploités en Guyane. Ces exploitations se font par des techniques gravitaires qui consistent à remettre en suspension les sédiments pour faire déposer les paillettes d'or qu'ils contiennent. Le lit majeur des cours d'eau, voire le lit mineur (après création d'un canal de dérivation), est exploité pour récupérer l'or alluvionnaire (Figure 21a). L'extraction du minerai s'effectue ensuite en délitant les couches de terrain au moyen de lances à eau à haute pression, avec ou sans l'appui d'une pelle hydraulique (Figure 21b). Le mélange de minerai et d'eau ainsi obtenu est canalisé vers un point de captage où une nouvelle pompe prend en charge le flux boueux pour l'envoyer vers des tables inclinées, ou l'or sera séparé des autres minéraux. Le flux boueux est dirigé dans des baranques, qui représentent des sortes de bassins de décantation permettant de limiter l'apport de particules fines dans l'eau des rivières. La réglementation en vigueur en Guyane impose l'utilisation d'un tel circuit fermé, cependant cette préconisation n'est pas toujours respectée par les orpailleurs légaux.et jamais par les orpailleurs clandestins.

L'orpaillage alluvionnaire se développe aussi de façon illégale en Guyane. Ces exploitations illégales rejettent de large quantité de boue directement dans la rivière exploitée et utilisent le mercure pour séparer l'or du reste des minéraux (Figure 21c). Cette activité clandestine crée des perturbations importantes, d'autant plus qu'elle s'est développée sur l'ensemble du territoire guyanais (Figure 20). Malgré les actions répressives des forces armées à partir des années 2010, le nombre de chantiers illégaux est en augmentation sur l'ensemble du territoire (Figure 22) et au moins 900 mines d'or à petite échelle ont été enregistrés au sein du bouclier guyanais alors que probablement beaucoup d'autres sont actuellement en cours de développement et non enregistrées (Hammond et al., 2007; Coppel et al., 2008).

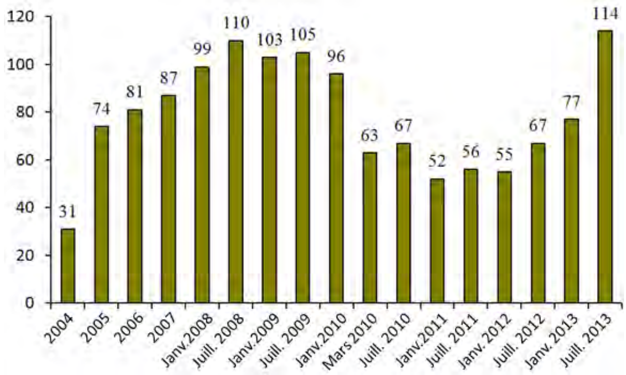


Figure 22 : Evolution du nombre de chantier illégal enregistrée en Guyane Française (Coppel et al., 2008)

I.2.3.2.3. Impact de l'exploitation aurifère sur les milieux aquatiques

Tous les principaux bassins, au sud comme au nord du département, sont actuellement touchés par l'activité aurifère. Ce constat souligne la grande nécessité d'outils de bioindication pour le suivi et la gestion de la ressource en eau du territoire. L'orpaillage modifie fortement la structure physique des habitats aquatiques au niveau des sites d'exploitation, et provoque des apports massifs de matière en suspension, augmentant aussi la turbidité de l'eau des rivières et des fleuves sur des dizaines de kilomètres en aval (Bruijnzeel, 1990 ; Dall'Agnol, 1995; Watts et al., 2003)(Figure 21d). Bien qu'une synthèse des valeurs de turbidité liée à l'orpaillage soit difficile à faire pour l'ensemble du territoire, des suivis réalisés par des organismes locaux (ex: Vigouroux et al., 2005, 2006) donnent des indications sur des valeurs de turbidité et MES dans des zones naturelles et impactées. Ainsi, en saison sèche, la turbidité naturelle qui est de l'ordre de 2-3 NTU (pour 0-7 mg/l de MES) et des valeurs ont été mesurées à 100-300 NTU (20-45 mg/l de MES) dans PME orpaillées. En saison des pluies, la turbidité naturelle est plus forte, de l'ordre de 10-15 NTU mais des valeurs d'autant plus extrêmes en milieu orpaillé ont déjà été mesuré dans des études et montent à 300-1500 NTU (Vigouroux *et al*,. 2005, 2006).

De très nombreuses études permettent de documenter l'impact biologique de l'augmentation des sédiments fins dans les rivières (Bruijnzeel, 1993; Parkhill et Gulliver, 2002 ; Utne-Palm, 2002 ; Krishnaswamy et al., 2006 ; Pekcan-Hekim et Lappalainen, 2006). De plus, en Guyane les relargages de sédiment sont suspectés d'avoir un fort impact sur les petits cours d'eau car ces milieux sont naturellement très faible en matières en suspension (Hammond et al., 2007). La plus faible pénétration de la lumière engendrée par les fortes turbidités entraine une diminution de l'activité photosynthétique, et par conséquent de la production primaire, affectant ainsi la biomasse des producteurs primaires et par la suite l'ensemble de la chaîne alimentaire (Davis et Simon, 1995, Tudesque et al., 2012.). La concentration excessive de matières en suspension peut diminuer la capacité de survie de certains poissons en diminuant l'efficacité de capture des chasseurs à vue et/ou en provoquant des abrasions et encrassements des branchies (Bruton, 1985). La sédimentation de ces matières en suspension peut aussi induire un colmatage du substrat et entrainer une diminution du nombre de micro habitats disponibles pour les macroinvertébrés benthiques ainsi qu'affecter le développement des diatomées (Richards et Bacon, 1994; Tudesque et al., 2012). De plus, l'orpaillage affecte indirectement les communautés aquatiques par l'ouverture du milieu provoquée par la coupe des arbres de la ripisylve. Il résulte de cette déforestation une augmentation de l'érosion des sols qui est une source supplémentaire de matière en suspension sur le long terme

(Wood et Armitage, 1997). Cela est due à la régénération très lente de la ripisylve sur les sites anciennement exploités comparé à d'autres types d'exploitation tel que « la coupe à blanc » (Peterson et Heemskerk, 2001). La disparition de la ripisylve provoque aussi une augmentation de la luminosité et de la température de l'eau, ainsi qu'une diminution des apports exogènes, de litière et d'embâcles (Bojsen et Barriga, 2002; Wright et Flecker, 2004).

I.3. CONSTRUCTION DE LA BASE DE DONNEES

I.3.1. Sélection des sites

La sélection des sites a été réalisée à partir d'une liste de sites potentiels sélectionnés sur dires d'experts grâce à une collaboration avec les partenaires de l'ONF et du PAG. Plusieurs critères ont orienté le choix des sites à échantillonner :

- Equilibrer l'effort d'échantillonnage entre les bassins versants pour couvrir l'ensemble du territoire et éviter d'éventuelles spécificités à un bassin. La majorité des sites échantillonnés se trouvent dans le rayon de la zone littorale. Cependant, plusieurs missions de terrain ont été effectuées pour permettre l'échantillonnage de sites situés plus à l'intérieur du pays. Ces sites, isolés de tout axe routier, ne sont accessibles qu'en se déplaçant en avion et/ou en pirogue, et nécessitent une lourde logistique. Certains de ces sites se trouvent en zone Cœur du parc ou en zone d'usage des populations amérindiennes et ont donc nécessité l'obtention d'autorisations auprès du PAG et des autorités coutumières. Le coût et les difficultés inhérentes à l'accès à ces sites isolés ne nous ont pas permis de réaliser une couverture aussi dense du territoire que dans la zone littorale.
- Réaliser des échantillonnages dans des sites de référence et des sites soumis aux principales perturbations anthropiques rencontrées sur le territoire Guyanais. Les principales activités humaines considérées sont de deux types : l'exploitation aurifère et forestière. L'ONF nous a fourni les droits d'accès aux zones d'exploitations forestières ainsi qu'un soutien logistique sur le terrain. Concernant l'orpaillage, il ne nous a pas été possible, pour des raisons évidentes de sécurité (l'essentiel des sites d'orpaillage sont des exploitations clandestines) d'accéder aux sites d'exploitation aurifère. Cependant, la turbidité de l'eau engendrée par l'activité d'orpaillage a été utilisée comme indicateur de perturbation, comme l'ont fait les travaux antérieurs (Vigouroux et al., 2005; Brosse et al., 2011). La sélection de sites anciennement orpaillés a été réalisée à partir de relevés de terrain et d'images aériennes de l'ONF et du PAG.

- Enfin, les sites sous influence de l'onde de marée ont systématiquement été exclus car ces zones ne sont pas considérées comme des petites masses d'eau mais comme des eaux de transition.

La deuxième étape de sélection consistait au choix du tronçon de cours d'eau. Les tronçons échantillonnés devaient se situer en amont de la piste lorsque celle-ci comportait un pont permettant le franchissement du cours d'eau, afin d'éviter les biais potentiels dus à l'impact de l'ouvrage. . Enfin, le tronçon devait aussi permettre l'échantillonnage sur une surface homogène et contigu. L'échantillonnage s'est déroulé durant les deux premières années (2011 et 2012) et a permis la collecte d'invertébrés aquatiques et une typologie physico-chimique dans 97 stations établies sur 93 rivières (Figure 23). Cette base de donnée est composée de 52 sites en condition de référence, 23 sites soumis à l'exploitation aurifère (actuellement et anciennement), 10 sites localisées en zone d'exploitation forestière et 12 sites soumis aux activités humaines (urbanisation, agriculture).

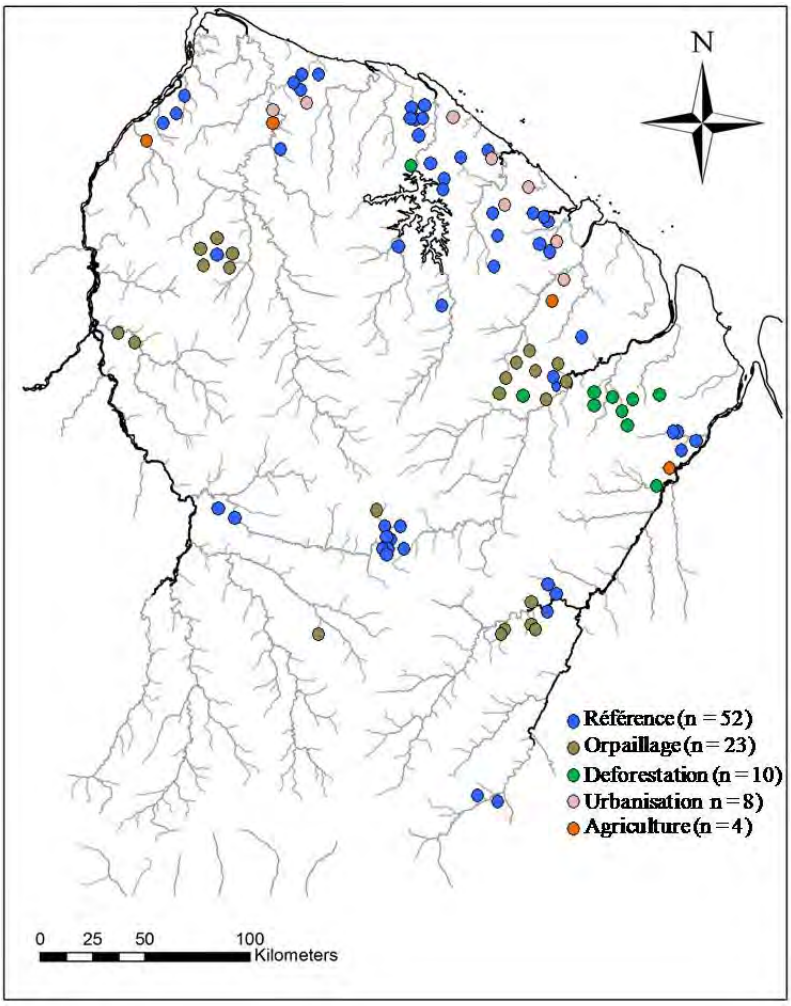


Figure 23 : Localisation de sites échantillonnés

I.3.2. Description des stations

Un protocole de description des conditions environnementales de la station a été mis en place. Celui-ci comporte deux étapes : les relevés de terrain et un travail cartographique.

Le travail cartographique, mené grâce au logiciel ArcGis sur les fonds de carte de la base BD Carthage, à partir de la localisation GPS de la station, permet d'accéder à des données relevées à l'échelle du bassin versant: distance à la source, pente et altitude de chaque station, ainsi que son appartenance à un bassin versant. Ces variables caractérisent les effets bassins et la position des sites dans le gradient amont-aval.

Les variables environnementales mesurées sur le terrain doivent permettre de rendre compte de l'effet des filtres régionaux et locaux sur les assemblages, c'est à dire des facteurs représentant l'habitat local :

- La largeur est mesurée sur au moins trois transects perpendiculaires à l'axe du cours d'eau, ou bien sur un transect tous les 5 mètres si la longueur de la station dépasse 10 mètres,
- La profondeur est mesurée tous les mètres le long des transects établis pour mesurer la largeur,
- Le pourcentage de recouvrement de chaque type de substrat : limon (substrat de taille inférieure à 0,05 mm), sable [0,05-2mm], gravier [2-10 mm], galet [10-30 mm], bloc [3-50 cm], dalle [>50 cm],
- Le pourcentage de recouvrement d'habitat organique de type embâcle, sous berge et racines, litière est visuellement estimé (des relevés ont été effectués sur chaque point où la profondeur a été mesurée, mais ces relevés ne sont pas significativement différents des estimations visuelles).
- Les caractéristiques chimiques de l'eau. Le pH, la conductivité, la concentration en oxygène dissous, la turbidité sont relevés sur chaque site à l'aide d'un PH-mètre WTW pH 3110 équipé d'une électrode WTW pH-SenTix 41, d'un conductimètre WTW Cond 3310 équipé d'un capteur tetraCon 325, d'un oxymètre WTW OXY 3250 et d'un turbidimètre Eutech Instruments TN-100. La température de l'eau, mesurée par plusieurs de ces appareils, a aussi été relevée.

Des prélèvements d'eau ont permis de mesurer la concentration en matière en suspension, le phosphore total et le nitrate. Ces prélèvements d'eau ont été analysés par le département chimie du laboratoire HYDRECO suivant des méthodes standardisées (AFNOR, 2000, 2005a, 2005b). Ces caractéristiques chimiques de l'eau permettent d'établir une typologie des masses d'eau et de prédire la présence de certaines perturbations, comme l'orpaillage (pour la turbidité et les matières en suspension (Vigouroux et al., 2005)) ou l'eutrophisation engendrée par les activités humaines (phosphore, nitrate - Smith et al., 1999).

Grace à ces différents paramètres, une caractérisation de la chimie de l'eau et des habitats physiques des petites masses d'eau guyanaise a ainsi pu être réalisée. Cela a en outre permis de caractériser les sites de références (Least Impacted River Reaches – LIRRS) et les sites perturbés (Impacted River Reaches - IRRS) au sein des différents hydro-écorégions (Tableau II).

Tableau II : Valeurs moyennes (Minimale – Maximale) des principaux paramètres environnementaux des sites de référence (LIRRS) et impactés (IRRS) dans les deux hydro-écorégions (Plaine littorale et Bouclier Guyanais).

Variables Habitat	**%Silt**	**%Sand**	**%Grav**	**%Coar**	**%Dw**	**%Macro**	**%Litt**	**%Root**	**%SubtratNue**
LIRRS									
Plaine littorale	8.03 (0 - 35)	61.87 (0 - 100)	23.66 (0 - 85)	6.87 (0 - 40)	16.33 (0 - 70)	10.44 (0 - 50)	24.01 (0 - 85)	20.25 (0 - 60)	48.21 (0 - 100)
Bouclier Guyanais	8.61 (0 - 25)	38.05 (0 - 95)	19.16 (0-55)	34.16 (0 - 85)	20.13 (0 - 65)	1.66 (0 - 15)	17.88 (2.5 - 60)	24.61 (0 - 70)	40 (0 - 100)
IRSS									
Plaine littorale	48.23 (20 - 90)	12.35 (0 - 55)	10.75 (0 - 35)	2.94 (0 - 15)	14.7 (0-40)	20.14 (0 - 65)	25.29 (0 - 50)	9.26 (0 - 27.5)	45.88 (5 - 100)
Bouclier Guyanais	34 (10 - 90)	15 (0 - 70)	19.66 (0 - 55)	32.66 (0 - 75)	7.33 (0 - 22.5)	1.66 (0 - 10)	18.83 (0 - 55)	7.83 (0 - 35)	74.33 (0 - 100)
Variables Eau	**ph**	**T**	**Cond**	**O2**	**Turb**	**NO3**	**PO4**	**TSM**	**DOC**
LIRRS									
Plaine littorale	5.29 (4.04 - 6.12)	24.75 (20.9 - 27.1)	22.25 (16 - 37)	6.01 (3.7 - 7.6)	2.06 (0.72 - 5.19)	0.30 (0.14 - 0.78)	0.057 (0 - 0.178)	6.32 (0 - 21.7)	22.03 (4 - 78)
Bouclier Guyanais	5.88 (4.18 - 6. 25	24.36 (19.9 - 28.4)	39.87 (21 - 114)	6.95 (5.6 - 7.9)	3.15 (1.07 - 11.02)	0.22 (0 - 0.52)	0.04 (0 - 0.103)	3.18 (0 - 7)	13.23 (0 - 38)
IRSS									
Plaine littorale	5.32 (4.7 - 5.8)	25.49 (24.2 - 27.9)	26.47 (12 - 60)	5,71 (4 - 7)	6.52 (0.51 - 31)	0,34 (0 - 0.7)	0.017 (0 - 0.063)	12.03 (0 - 48.8)	20.31 (7 - 47)
Bouclier Guyanais	6.38 (5.71 - 6.87)	25.64 (23.8 - 30.7)	48.93 (25 - 135)	6.84 (5.1 - 7.7)	54.69 (2.5 - 329)	0.298 (0.11 - 0.51)	0.024 (0 - 0.08)	58.79 (0 - 566)	12.26 (0 - 21)

I.3.3. Echantillonnage de la faune benthique

Afin d'effectuer des échantillonnages sur le plus grand nombre de sites, la période de terrain est restreinte à la saison sèche. Ce choix a permis d'effectuer des échantillonnages dans des conditions hydrologiques stables et comparables. Le petit été de mars étant trop variable, la saison sèche (mi-septembre à mi-décembre) est préconisée pour réaliser les collectes d'invertébrés. Les prélèvements ont été réalisés à l'aide d'un troubleau carré de 20 centimètre muni d'un filet de 0,5 millimètre de vide de maille. De l'alcool à 70% et des bocaux avec des opercules sont utilisés pour la conservation des insectes. Le nombre de prélèvements a été fixé à 12 :

- 8 prélèvements unitaires des habitats organiques, généralement situés sur la berge (Phase A). Ce prélèvement se déroule selon un temps de **30 secondes** en effectuant mécaniquement des 8 avec le filet troubleau,
- 4 prélèvements unitaires des habitats minéraux, généralement situés dans le chenal (Phase B). Le prélèvement des substrats minéraux se fait sur une surface. Pour être indicative, cette surface de prélèvement doit dépendre de la taille du cours d'eau. Dans notre cas, les petites

masses d'eaux variant peu (2-10m de largeur), la surface de prélèvement est comprise entre **0,05 et 0,75 m^2**.

Le prélèvement d'un substrat, pour l'inclure dans un protocole représentatif et comparable, doit représenter une surface minimale au moins égale à un pour mille de la surface de la station (estimation visuelle). Les sélections des types de substrats organiques (Phase A) et minéraux (Phase B) ont été fait en fonction de leur habitabilité (capacité du substrat à accueillir une faune diverse - Tableau III). Les prélèvements sur un substrat récurrent ont été faits sur différentes vitesses de courant.

Tableau III. Définition et ordre d'échantillonnage des substrats (Hydreco, Unpublished data)

Type de substrat	Définition	Habitabilité
Substrats organiques (Phase A)		
Salade Coumarou	Uniquement dans les sauts (fleuves)	10
Système Racinaire	Chevelus racinaires libres (surface)	9
Tapis Racinaire	Supports ligneux.	8
Macrophytes	Spermaphytes immergés (Hydrophytes)	7
Moucous-Moucous	Uniquement en bordure de fleuve	6
Végétation terrestre	Spermaphytes émergents (Hélophytes)	5
Litière	Débris organiques grossiers (feuilles)	4
Bryophytes	Tapis de mousse sur support minéral ou organique.	3
Limon/Vase	Sédiments et débris organiques fins	2
Latérite Nue	Berge érodée principalement.	1
Substrats minéraux (Phase B)		
Galet	Sédiments minéraux de grande taille (25 à 250 mm)	5
Gravier	Granulat grossier (2 à 25 mm)	4
Bloc	Eléments minéraux de grande taille (>250 mm)	3
Sable	Sédiments fins (< 2mm)	2
Dalle/Roche	Surface Uniforme plate	1

I.3.4. Les invertébrés récoltés

Au total, 10654 individus ont été prélevés. Les invertébrés capturés sont majoritairement des arthropodes (9 ordres), mais aussi des crustacés, annélides et des mollusques (Annexe I). Au total 95 familles ont été recensées au cours de cette étude. Chaque site comprenait de 8 à 63 familles (Tableau IV). L'abondance des invertébrés capturés par site était très variable (de 70 à 6135 individus).

Tableau IV : Les richesses, abondances et pourcentages moyens (minimaux – maximaux) des principaux ordres d'invertébrés benthiques récoltés au cours des deux campagnes d'échantillonnages.

	Année 2011	Année 2012
Richesse taxonomique	37.18 (8 - 63)	40.95 (18 - 51)
Abondance	1690.18 (70 - 4519)	1475.88 (263 - 6135)
% Diptères	46.80 (10 - 95.31)	58.09 (25.88 - 81.07)
% Ephéméroptères	14.58 (0 - 45.23)	10.19 (3.39 - 24.66)
% Trichoptères	9.52 (0.43 - 72.85)	10.06 (1.73 - 28.63)
% Coléoptères	6.98 (0 - 25.56)	9.60 (0.15 - 41.44)
% Odonates	2.61 (0 - 9.51)	2.27 (0.38 - 6.71)

I.3.5. Composition des bases de données utilisées dans les différents chapitres (Tableau V)

Les chapitres II, III et IV sont complémentaires et ont permis d'élaborer les différentes bases nécessaires pour la construction d'un outil de bioindication (caractérisation des sites de référence, prise en compte de la variabilité naturelle, élaboration de l'indice). Le chapitre V est une étude préliminaire qui a pour objectif d'évaluer la capacité bioindicatrice d'un groupe en particulier : les Ephéméroptères. Le nombre de site est réduit en raison du travail supplémentaire d'identification requis (niveau générique).

Tableau V: Nombres de sites, types de données et les analyses statistiques utilisées dans les différents chapitres de la thèse.

Chapitres	N sites (LIRRs/IRRs)	Données utilisées	Analyses statistiques
Chapitre II. Impact de l'orpaillage et de la déforestation sur la chimie de l'eau et les habitats physiques des petits cours d'eau de Guyane Française	57* / 38	Variables environnementales (19 variables)	Comparaisons de moyennes (Kruskal Wallis - KW) Multivariées (Analyses de correspondance principale (ACP))
Chapitre III. Les communautés d'invertébrés délimitent des hydroécorégions et répondent aux perturbations dans les cours d'eau Est-Amazoniens	40 / 25	Variables environnementales (22 variables) Biologie (86 Taxons, niveau familial)	Comparaisons de moyennes (KW) Analyse auto-organisatrice (réseau de neuronne)
Chapitre IV: Un indice multimétrique basé sur les invertébrés pour la mise oeuvre de la directive cadre européenne sur l'eau en Guyane Française	52 / 45	Variables environnementales (17 variables) Biologie (95 T., n. familial)	Comparaisons de moyennes (KW) Test d'hypothèse (Kolmogorov-Smirnov)
Chapitre V : Evaluation de l'impact de l'orpaillage sur les cours d'eau de tête de bassin de Guyane à l'aide des traits biologiques des éphémères	6 / 7	Variables environnementales (14) Biologie (35 T., d'Ephéméroptères, n. générique)	Comparaisons de moyennes (KW) - Analyse de variance multivariée (MANOVA) Multivariées ((ACP, analyse factorielle des correspondances avec codage flou)

** 5 sites supplémentaires échantillonnés au cours d'un autre projet ont été utilisés (Allard et al., 2014).*

Chapitre II. Impact de l’orpaillage et de la déforestation sur la chimie de l’eau et les habitat physique des petits cours d’eau de Guyane Française.

Ce chapitre a fait l’objet d’une publication dans la revue scientifique Knowledge and management of aquatic ecosystems.

Résumé

La gestion des écosystèmes aquatiques nécessite des systèmes d'évaluation prenant en compte la typologie des cours d’eau, ce qui permet de considérer la variabilité naturelle et d’optimiser l’efficacité des programmes de surveillance. Cette approche est exigée par la directive Cadre Européenne de l’Eau. Nous avons établi une typologie des petits cours d'eau de Guyane Française en utilisant des critères physico-chimiques, définissant ainsi des contextes environnementaux pour l’analyse future des communautés biologiques. Les petits cours d’eau de Guyane Française représentent 70 % du réseau hydrographique du département, et ces milieux sont localement soumis à de fortes pressions (déforestation et orpaillage). Nous avons émis l’hypothèse que l’exploitation aurifère et forestière affecte principalement la structure physique de l'habitat, et que ces deux types d’exploitation ont une influence sur le compartiment chimique par la modification du flux de nutriments et/ou la remise en suspension de particules fines.

95 sites répartis en quatre catégories de perturbation (référence, exploitation forestière, orpaillage actuel et ancien) ont été caractérisés par des variables physico-chimiques. Nous avons démontré que les variables physiques décrivant le lit du cours d’eau et les matières en suspension différencient les sites soumis à l’exploitation aurifère et forestière des sites de références. Les concentrations en nutriments ne sont pas significativement modifiées par les impacts humains. Ces résultats mettent en évidence l'effet négatif et persistant de l'exploitation aurifère sur l'habitat physique des rivières alors que les effets liés à la déforestation semblent moins sévères. Cela est probablement en lien avec les actions de protection en vigueur en Guyane. Cependant, les tendances de l'exploitation aurifère (principalement illégale) vers les zones amont ou les cours d’eau présentent encore de très bonnes qualités écologiques soulignent le besoin de construire des méthodes de bioindication efficace. Parce que les organismes sont de parfaits intégrateurs de la structure et du fonctionnement de leur milieu, nous pouvons nous s'attendre à une diminution de la qualité biologique des zones amont. Cette typologie préliminaire sera donc pertinente par la suite pour étudier les patrons de distribution de la faune aquatique, et pour construire un outil de biosurveillance des cours d'eau.

Physical habitat and water chemistry changes induced by logging and gold mining in French Guiana streams

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Keywords: Neotropical steams; headwaters; reference conditions; deforestation; gold mining

Abstract

Understanding the effects of disturbances on the physical-chemical quality of ecosystems is a crucial step to the development of ecosystem assessment tools. 95 sampling sites distributed among 4 categories of disturbance, i.e.: reference, logging, formerly and currently gold mining, were characterized using stream physical and chemical variables. Our hypotheses were: (i) logging and gold mining activities primarily affect the physical habitat structure of streams and (ii) both have an effect on chemical environments through nutrient and/or fine particulate resuspension. We demonstrate that physical variables describing the river bottom, and suspended solids discriminate both current and formerly gold mined sites from reference sites, while, whatever the type of impact encountered, nutrient concentrations do not prove relevant to measure human impacts. To understand distribution patterns of aquatic organism across FG, future research should thus aim at examining the match between physical-chemical and biological classifications of small streams under reference and impacted conditions.

II.1. INTRODUCTION

Under most water management policies, ecosystem health is defined in terms of similarity to a near-pristine, undisturbed state (Bailey et al., 2003). In practice, predictions of the physical-chemical and/or biological conditions to be expected under undisturbed conditions in any given geographic area are based on the classification of river sites. By knowing what set of environmental conditions should be encountered at undisturbed (or least impacted) sites, one can then estimate the degree to which impacted sites are altered by human activity (Bennett et al., 2011). In addition, physical-chemical classifications of rivers provide a template against which changes in biological diversity within watersheds can be interpreted in relation to natural variability and anthropogenic impacts (Van Sickle and Hughes, 2000).

Recent studies in temperate areas have prompted a large amount of characterizations of reference and impacted physical-chemical environments (e.g., Tudesque et al., 2008). However, differences in bioclimatic, biogeographic and geomorphological conditions preclude the transposition of current typological schemes to EU's overseas regions (see Touron-Poncet et al., 2014 for a rationale), while limited scientific effort has been directed at typifying rivers in overseas Europe in terms of physical-chemical (and biological) patterns. Therefore, as a prerequisite to any methodological development, there is a pressing need to collect environmental information in a standardized manner so that fundamental data can be analyzed in an integrated way.

French Guiana (FG) is an overseas region of France located on the northern coast of South America. About 96% of its surface area (over 82 000 km^2) is covered by equatorial forest, which partly belongs to a recently-created National Park. The Guianese primary forest remains one of the least impacted of the World, however, gold mining and timber have strong impacts upon river ecosystems. Specifically, the annual gold output in the area is 60 times higher than 25 years ago (Hammond et al., 2007). After the prospection of large rivers, gold industries are now focusing on smaller inland streams (Cleary, 1990; Hammond et al., 2007). Small streams (from headwaters to rivers with depth < 1m and width < 10m) represent 70% of all running waters in FG. Most of small streams are located in forested areas and exhibit high ecological quality; some if not most of them have never been impacted by any human activity. In light of recent economic development, our ability to predict both reference conditions and ecosystem responses to landscape alterations will determine the success of future management actions.

We relied on extensive characterizations of stream physical (particle size, substratum heterogeneity) and chemical conditions (e.g., nitrogen, phosphorus) at 95 sampling sites distributed over 95 streams and representing 4 categories of disturbance: reference (unimpacted), deforestation, ancient gold mining, and ongoing gold mining. First, we tested if impacted sites are randomly located within the river continuum or if they are characterized by particular local physical features that distinguish them from the references. Our hypothesis was that, at any given location within a stream system, gold mining and deforestation primarily affect the physical habitat structure and/or heterogeneity. Second, we tested differences in chemical variables among site categories. Assuming that both deforestation and gold mining affect chemical environments through nutrient and/or fine particulate runoff/resuspension, we expected that sediment resuspension due to gold mining would result in harsher shifts in instream environmental conditions.

II.2. MATERIAL AND METHODS

II.2.1. Study area

This study was conducted in French Guiana (surface area = 83,534 km^2), East Amazonia, from September 2011 to December 2012. The climate is tropical moist with 3,000 – 3,400 mm of yearly precipitation mainly distributed over 280 days. There is a major drop in rainfall (dry season) between September and December and another shorter and more irregular dry period in March. The maximum monthly temperature averages 33.5 °C (32.1-35.8 °C), and the monthly minimum averages 20.3 °C (19.7-21 °C). French Guiana's stream systems are organized around seven large rivers (Maroni, Kourou, Mana, Sinnamary, Comté, Approuagues, and Oyapock rivers); however, the "small streams" sampled in this study (water depth < 1m; stream width < 10m) represent ca. 80 000 km in total length, i.e. 70% of all running waters in the region. We did not considered larger streams and rivers to focus on comparable ecosystems located in the upstream part of the river continuum.

Figure 24 : Map of French Guiana showing the main rivers and the location of the sampling sites.

II.2.2. Sampling sites and environmental variables

Our 95 sampling sites were distributed over 95 small streams belonging to FG's main river basins (Figure 24). It should be noted however that the sampling effort was higher in the Northern part of FG, due to the difficulty to access southern FG. In this specific area covered by dense rainforest and without road networks, complex logistics limited our ability to sample a larger number of sites. We thus managed to collect some samples from the main southern river basins (Figure 24). All sites were sampled during the dry season in 2011 and 2012 (September-December). Indeed, pollution is detected less efficiently during high flows because of dilution. In addition, most remote sites cannot be reached (and therefore monitored) during the rainy season. Each site was sampled once, and the sites sampled in 2012 are hence distinct from those sampled in 2011.

Based on expert knowledge and field observations, sampling sites were categorized into four a priori groups corresponding to four types of anthropogenic pressure. Reference sites (Ref, n=57) were defined as sites not subjected to anthropogenic impacts such as gold mining, deforestation, chemical pollution, agricultural or urban runoff. Deforested sites were subjected to logging for wood products and timber (Log, n=15), under the supervision of the National Forest Office (ONF). The ONF manages a sustainable logging industry based on strict plans intended to minimize impacts on the environment. The remaining sites were formerly subjected to gold mining but no longer exploited (Fog, abandoned mining n=9), or currently subject to gold mining (Cug, n=14). Either formerly or currently gold mined sites refer to illegal gold mining activity. "Illegal" mining refers here to so-called "informal" mining, i.e., small-scale traditional (or artisanal) mining which also occurs in most South American countries (Hammond et al., 2007). Mercury is used for gold amalgamation during the mining process, and about 30% of the mercury is released into the river. Mercury concentration in the water column is however insignificant (see Coquery et al., 2003, Maury-Brachet et al., 2006 for documented cases in FG). In addition, the effects of heavy metals on the composition of biological communities expected to form Biological Quality Elements in subsequent developments are not obvious (De Jonge et al., 2008). Although mercury was not taken into account in our study, it should be noted however that river sediments have a strong adsorption capacity for heavy metals (Pfeiffer et al., 1989; Roulet et al., 1999), and may certainly be a relevant parameter to quantify the impact of gold mining. Mercury can subsequently accumulate in plant and animal tissues before entering food chains – in French Guiana some concentrations in edible parts of locally consumed fish can surpass the advisory level for human consumption, thus forming a key concern for human health (Durrieu et al., 2005).

Stream scale variables, namely elevation above sea level, distance from the source, and slope were obtained from Geographic Information System (GIS, ESRI ArcGis 10). These variables characterise the location of sampling sites within the upstream-downstream river continuum. Site scale variables were chosen to describe the heterogeneity of riverbed substrate and habitat availability at each site. They were recorded directly in the field and accounted for the percentage composition of organic and mineral substrate types, using the standardized protocol by Souchon *et al*., (2000). These variables included: %leaf litter (Litt), %submerged roots on the banks (Bank), %submerged vegetation, mostly macrophytes (Macr), %woody debris (Wood), %Silt, %Sand (particle size < 2mm), %Gravel (Grav 2-25mm), %coarse substratum (Coar > 25mm). Coarse substrates being scarce in French Guiana, this category of mineral substrate included pebbles, boulders, and/or rocky outcrops. In addition, we recorded the stream width (Widt, m) and water depth (Dept, m). The forest canopy coverage (Fore) was evaluated visually, from 0 to 100% (Table VI). We also measured chemical variables accounting for the chemical impairment of stream ecosystem (PO_4^{3-} and NO_3^-) by human activities and for the transport of solids (Total Suspended Solid and Turbidity) (Table VII). Turbidity was measured directly in the field using an Eutech Instruments Turbidimeter (TN-100). Other chemical analyses were carried out at Hydreco Laboratory, Petit-Saut, based on water samples taken at each site and immediately frozen. Chemical analyses followed standard methods summarized in AFNOR (2000-2005).

Table VII: Chemical variables used to assess human disturbance on the four streams category (Ref : Reference; Log; Logging; Fog: Formerly gold mined; Cug: Current gold mined). Values indicate mean± SD.

Chemical variables	**unit**	**Ref**	**Log**	**Fog**	**Cug**
Turbidity	*NTU*	5.31±22.5	5.51±11.1	32.5±93.4	27.6±43.9
$NO3^-$	*mg/L*	0.31±0.16	0.25±0.21	0.34±0.08	0.25±0.15
PO_4^{3-}	*mg/L*	0.04±0.02	0.04±0.05	0.02±0.02	0.02±0.02
Total Suspended Solid	*mg/L*	6.1±12.8	8.4±12.9	9.9±14.4	60.4±155.4

Tableau VI: Environmental variables used to describe the four streams category (Ref : Reference; Log; Logging; Fog: Formerly gold mined; Cug: Current gold mined). Values indicate mean± SD.

	code	unit	Ref	Log	Fog	Cug
Stream scale variables						
Elevation	Elev	m a.s.l.	131.3±153.7	57.2±18.1	80.1±36.3	90.8±24.9
Distance from headwater source	Dist	km	3.3±3.8	1.9±2.4	3.6±2.6	5.61±3.4
Slope	Slop	‰	4.7±3.6	4.7±3.4	4.7±3.19	3.84±2.6
Site scale variables						
Bank	Bank	%	12.1±13.2	14.1±12.1	9.2±7.2	7.79±5.4
Macrophyte	Macr	%	3.1±17.4	3.7±13.7	2.2±6.5	0.83±2.7
Litter	Litt	%	24.1±19.8	24.1±23.9	16.7±19.8	25.5±31.3
Woody debris	Wood	%	14.1±12.9	20.9±22.3	13.2±11.6	7.26±7.9
Silt	Silt	%	11.9±18.1	16.3±17.5	21.7±12.3	26.2±27.7
Sand	Sand	%	47.2±31.9	49.5±24.7	26±28.17	16.4±20.4
Gravel	Grav	%	20.7±22.9	11.4±15.4	35.7±27.8	30.7±31.6
Coarse substratum	Coar	%	19.6±25.9	12.7±15.4	26.6±25.5	26.7±27.8
Width	Widt	cm	379.9±231.5	429.3±271.2	393.6±198.7	394.3±187.5
Depth	Dept	cm	25.5±11.9	26.7±14.2	23.1±7.9	27.9±13.3
Forest Coverage	Fore	%	74.8±16.3	55.1±28.3	57±27.4	64.6±23.6

II.2.3. Data analysis

We first used a Principal Component Analysis (PCA) to ordinate the sites according to topological and physical variables, and to bring out potential shifts in physical conditions following anthropogenic disturbance. Prior to analysis, continuous variables were log-transformed, and discrete variables expressed in percentages were Arcsin-transformed. Plots of the first two ordination axes usually capture most of the variance and consequently contain most of the information that is likely to be interpretable (Waite et al., 2000). Neighbouring sites in the scatterplots were expected to define areas with similar physical environments. Conversely, sites having a large distance to each other were expected to be distant in the feature space, according to environmental characterization. In order to compare distributions of sites according to disturbance types, a Kruskal-Wallis (KW) test was performed on the site coordinates of the two first axes of the PCA. To further bring out relationships between water chemistry, local environments and disturbance, significant differences among a priori groups were also tested using Kruskal-Wallis tests on the raw values of measured parameters. Then, significant differences in physical characteristic between a priori groups of sites (Ref, Log, Fog, Cug) were further assessed using Wilcoxon tests.

As different sites were sampled in 2011 and 2012, the sampling year was not informative and we hence pooled the two years data. All computations were performed using the R

Software (R Development Core Team, 2003), the ADE-4 (Thioulouse et al., 2007) and Vegan (Oksanen et al., 2013) package.

II.3. RESULTS

Eigenvalues for axis 1 and 2 of the PCA were 3.43 and 2.26, respectively (Figure 25a). The first and second axes explained 24.53% and 16.7% of the overall variance, respectively. The distribution of sampling sites in the scatterplot did not show clear clumps according to environmental characteristics, but rather displayed a predictable, upstream to downstream gradient. Axis 1 thus displayed a gradient of elevation, slope and substratum size (from high (left) to low (right)). These parameters are related to the river competence (i.e. the maximum size or weight of material a river can transport). Axis 2 accounted for stream width and depth, and distance from source (from high (upper area) to low (down)).

Sites subjected to current and former gold mining were mostly distributed along axis 2, while sites subjected to logging were distributed along axis 1. Only reference sites and sites subjected to current gold mining differed significantly in their distribution along axis 1 (Figure 25b), currently exploited sites being more concentrated in the upstream areas. When the distribution of sites was examined along axis 2 (Figure 25c), both formerly and currently gold mined sites differed from other sites, while reference sites and sites subjected to logging did not show significantly different distributions.

Stream scale variables showed significant differences between impairment categories. The mined sites had coarser substrates than the references and logged sites, as shown by a significant difference in the percentage composition of mineral particles. Significant differences were found in %sand (KW-chi-square = 15.3776, df = 3, p-value = 0.001521), %silt (KW-chi-square=12.2781, df = 3, p-value = 0.006489) and %gravel (KW-chi-square=7.6945, df = 3, p-value = 0.04277). Such a difference between reference and mined sites holds true for both the formerly and currently gold mined sites.

Considering chemical variables, neither PO_4^{3-} and NO_3^- (Figure 26a & 26b), nor suspended solids and turbidity (Figure 26c & 26d) showed significant differences between reference sites and sites subjected to logging. Gold mining did not alter PO_4^{3-} and NO_3^- concentrations (Figure 3a & 3b), however, stream turbidity values (Figure 26d) were significantly different from values observed at reference and logged sites (see appendix II for outputs of Wilcoxon tests).

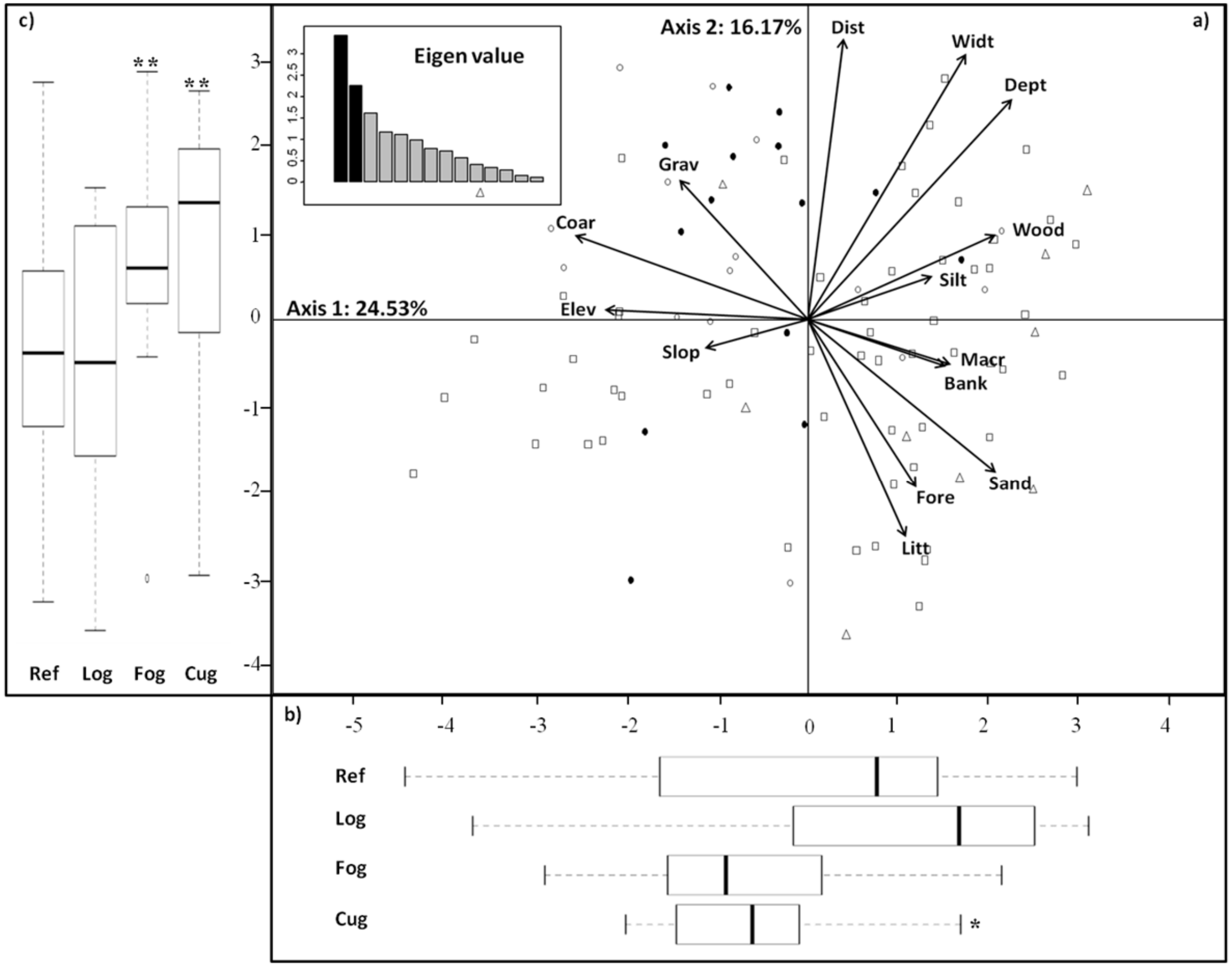

Figure 25: (a) Principal Component Analysis (PCA) biplot showing the distribution of the 95 sites according to environmental variables (See table VI for acronyms). Rectangles: reference sites; triangles: logged sites, open circles: formerly gold mined sites and filled circles: currently gold mined sites. (b) Boxplots of the coordinates of the sites on the first axis. (c) Boxplots of coordinates of the sites on the second axis. Ref: Reference Sites; Log: Logged Sites; Fog: Formerly gold mined sites; Cug: Currently gold mined sites.
(: P-value <0.1; ***: P-value <0.01).*

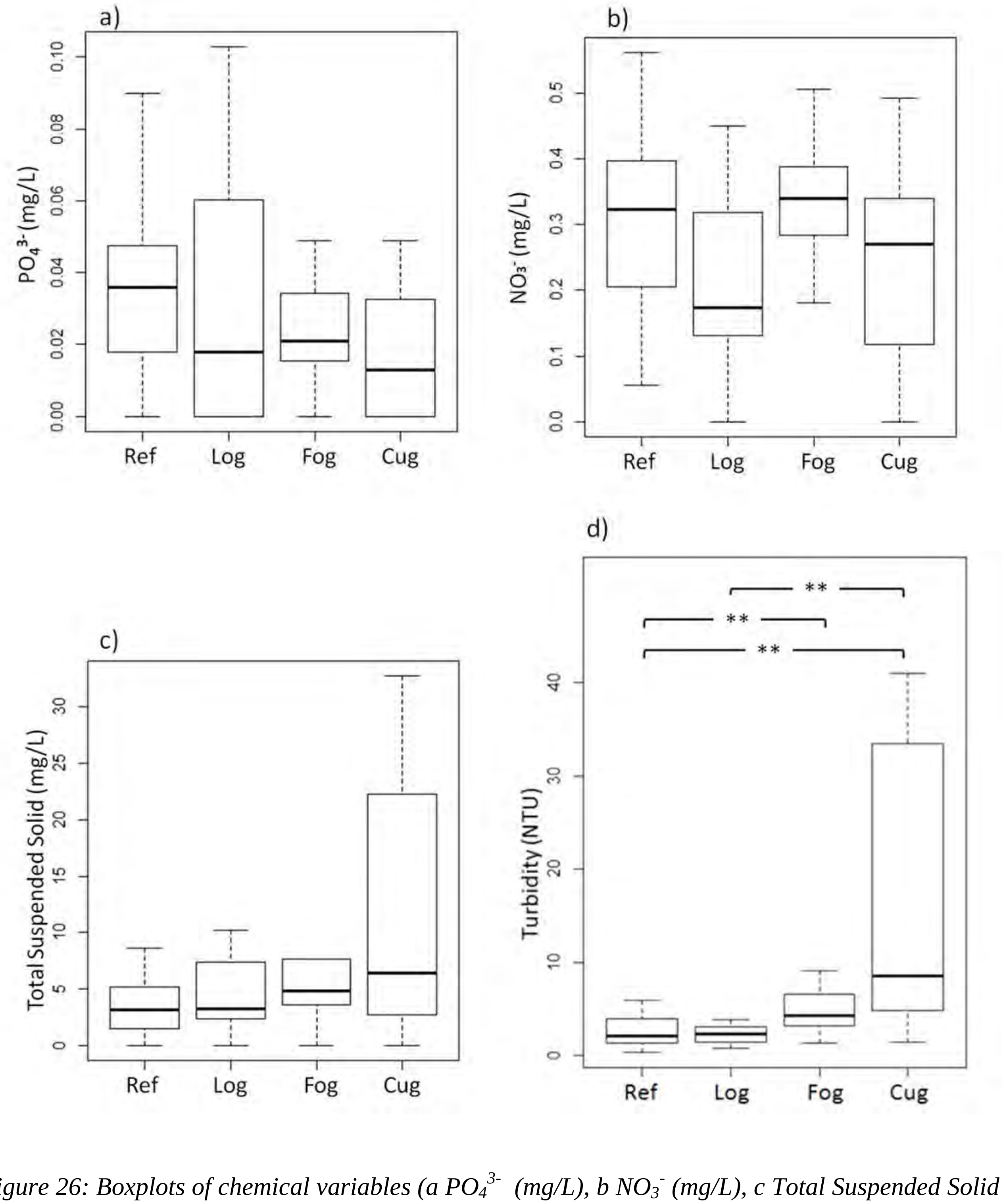

*Figure 26: Boxplots of chemical variables (a PO_4^{3-} (mg/L), b NO_3^- (mg/L), c Total Suspended Solid (mg/L) and d Turbidity (NTU)) in reference (Ref), logged (Log), formerly goldmined (Fog) and currently goldmined (Cug) sites. Limits of the box represent the first and third quartiles, bold line is the median, and whiskers are extreme values. Stars indicate the significance of Kruskal Wallis test between classes (**: P-value <0.05).*

II.4. DISCUSSION

Given the contrasted types of human activities that affect stream ecosystems in FG, we expected significant differences in physical-chemical characteristics of impaired streams in relation to disturbance type.Sites subjected to logging (deforestation) had finer bottom substrates than the reference sites. This can be related to increased inputs and deposits of fine particles, brought to the stream through tractor tracks and gravel road creation for logging trucks (Forman and Alexander, 1998). Such a tendency is triggered under equatorial climate, where harsh rains (rainy season) have a strong erosive effect on bare lands (Dudgeon, 2008). Despite increased siltation, suspended solids and water turbidity were not affected, highlighting the moderate effect of logging on streams in FG.

It should be noted however that logging is strictly controlled in FG, in order to minimise environmental impacts. Specific measures include the absence of clear-cutting, and the protection of the riparian zone where logging is forbidden. In the same way, logging trucks cannot be used during the rainy season and cannot cross streambeds, thus reducing sediment load to the aquatic ecosystems (Panchout, 2010). Such management efforts seem to prove efficient, as we did not detect any significant effect on turbidity and other chemical variables that are usually strongly sensitive to intensive logging activities. Specifically, leaching of soils and canopy opening are known to modify nitrogen and phosphorus fluxes to the aquatic ecosystem (Sweeney et al., 2004; Neill et al., 2006).

The river bottom of most gold mined sites was characterised by a dominance of gravels. This trend is not related to natural processes. Gold deposits are collected by washing the soils adjacent to the streams with high pressure water jets, and gravels are then sieved and released to the stream (Hinton et al., 2003), therefore increasing their prevalence over the streambed. During these operations, the streams also receive the draining water that contains a high load of sediments (Watts et al., 2003), explaining the higher turbidity at gold mined sites (see also Mol and Ouboter, 2004; Brosse et al., 2011).

Contrary to our expectation that gold mining, through the predicted clearing of the riparian forest and soil leaching, should increase stream eutrophication (see Hammond et al., 2007; Palmer et al., 2010), we did not find any significant change in nutrient loads (PO_4^{3-} and $NO3^-$) in gold mined sites. This result can be explained by the illegal nature of the exploited sites, which typically remain hidden under the canopy and do not host more than 30-40 workers (Hinton et al., 2003). There is therefore no deforestation at these sites, and hence no drastic shift in nutrient fluxes.

This however does not mean that the ecosystem is not impaired. In particular, it has been demonstrated that both fish and diatom assemblages are strongly affected by the turbidity generated by small scale gold mining through habitat clogging and changes in light penetration over the bottom (Cleary, 1990; Mol and Ouboter, 2004; Brosse et al., 2011; Tudesque et al., 2012).

It is worth noting that turbidity remained significantly higher in formerly gold mined sites than in reference sites. This is probably due to fine sediment storage in the stream pools, so that these sediments can be re-suspended in the river column when river discharge increase during the frequent rain events. Therefore, temporary physical disturbances of stream ecosystems can persist in time, explaining why biological assemblages do not recover after stopping mining (e.g., fish and diatom, see Brosse et al., 2011; Tudesque et al., 2012).

Finally, the comparison of formerly and currently gold mined sites revealed a shift of activities towards the upstream sites. This might be afforded to two non-mutually exclusive reasons. First, the rarefaction of the gold resources as well as the rise of gold prices brought gold miners to move deeper in the forest (Cleary 1990; Hammond et al., 2007) and exploit more remote upstream sites. Second, the increased control of illegal gold mining by French authorities (Coppel et al., 1998) forces illegal miners to exploit those remote sites and to remain as inconspicuous as possible. This probably explains why in most of our sites we did not observe deforestation that would make the mining sites easily detected (Hinton et al., 2003).

In conclusion, we demonstrated that, under unimpacted conditions, there is no clear clustering of freshwater streams in French Guiana, thus complicating aims to set up a stream typology based on physical characteristics. Also, whatever the type of impact encountered in small streams of FG, nutrient concentrations did not prove relevant to measure human impacts. Logging did not result in detectable impacts on stream physical characteristics, probably because this type of activity is strictly managed and controlled by local stakeholders. However, site scale variables that describe the riverbed, habitat and suspended solids (i.e. simple physical measurements) clearly segregated both currently and formerly gold mined sites from reference sites. These results highlight the persisting, adverse effect of mining on the benthic habitat. Assuming that the structure of biological communities in streams are not due to random processes (Minshall and Petersen, 1985) but is strongly influenced by physical factors such as stream bed morphology (Wallace and Webster, 1996), hydrological conditions (Power et al., 1988), one can assume that substrate homogenization by anthropogenic activities will largely constraint the benthic community structure. Moreover, invertebrates or diatoms are tightly integrated into the structure and functioning of the benthic ecosystem, one may expect dramatic decreases in the biological quality of headwater streams with the shift of gold mining towards the upstream areas. To design potential biological indication tools of impairment, future research should thus aim at examining the match between physical-chemical and biological classifications of small streams under reference and impacted conditions.

Chapitre III. Les communautés d'invertébrés délimitent des hydro-écorégions et répondent aux perturbations dans les cours d'eau Est-Amazoniens .

Ce chapitre a fait l'objet d'une publication dans la revue scientifique Hydrobiologia.

Résumé

En Europe, la Directive Cadre sur l’Eau implique que les outils d’évaluation de la santé des écosystèmes aquatiques développés au sein des états membres répondent à un certain nombre de critères communs. Deux critères sont particulièrement importants et applicables à l'ensemble des éléments de qualité biologique en eau courante: (i) l'obligation de réaliser l'évaluation par rapport à une situation de référence représentant l’état « naturel » et (ii) une évaluation qui doit prendre en compte la typologie systèmes étudiés. Aucun modèle cartographiant les conditions biologiques et environnementales n’existait à ce jour pour la Guyane Française, limitant donc la mise en œuvre de la DCE sur la base de l'état référence. En nous concentrant sur les petits cours d’eau de Guyane (profondeur <1m, largeur du lit <10m ; 70% des cours du réseau hydrographique) nous avons testé deux prédictions : (i) la géomorphologie détermine des sous-régions écologiques qui ont des communautés d’invertébrés homogènes, et (ii) la diversité des communautés diminue alors que les pressions anthropiques augmentent.

65 sites ont été caractérisés par leurs compositions en invertébrés benthiques et des variables physico-chimiques dans différents bassins versants. Nous avons utilisé un réseau de neurones non supervisé (self organizing maps, SOM) afin de mettre en évidence les relations entre les communautés d'invertébrés et les variables environnementales. Les sites caractérisés par les communautés d'invertébrés se sont regroupés en deux grandes sous-régions correspondant aux hydro-écorégions de la Guyane française: « la plaine alluviale » caractérisée par des sédiments récents et une faible altitude, et le « bouclier guyanais » caractérisé par un substrat rocheux et un couvert forestier dense. Les changements dans la composition de la communauté, et dans une moindre mesure la richesse taxonomique au sein de chaque sous-région ont révélées un impact lié aux exploitations aurifères et forestières, découpant à nouveau les hydro-écorégions en deux sous-ensembles, les sites de référence et les sites impactés. Des analyses supplémentaires sont toutefois nécessaires pour quantifier les écarts écologiques entre les états naturels et perturbés, en particulier au niveau des têtes de bassins où l’orpaillage a un impact si sévère que les communautés d’invertébrés benthiques y sont comparables à celles des sites les plus impactés des zones aval.

Invertebrate communities delineate hydro-ecoregions and respond to anthropogenic disturbance in East-Amazonian streams

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Abstract

Many tropical regions lack models predicting the biological and environmental conditions expected in any given area, thus precluding the implementation of reference condition-based water policies. We focused on streams of French Guiana, and tested two predictions: geomorphology determines ecological sub-regions that have typical invertebrate communities, and diversity declines as anthropogenic pressure increases. Sixty-five stream sites were sampled for benthic invertebrates and physical-chemical variables across various watersheds. We used the Self-Organizing Map algorithm (neural network) to model relationships between invertebrate communities and environmental variables. Sites characterized by invertebrate communities clustered into two major subsets matching French Guiana's hydro-ecoregions: the coastal alluvial plain characterized by recent sediment and low elevations, and the Guiana Shield characterized by an eroded rocky substrate and dense rainforests. Changes in community composition, and to a lesser extent taxonomic richness within each sub-region revealed ecological impacts of gold mining and logging, further clustering hydro-ecoregions into subsets of reference and impaired sites. Further analyses would however be needed to identify tipping points between natural and disturbed states, especially in remote headwater streams where gold-mining had the harsher impact upon freshwater diversity, making upstream communities resembling the most downstream impacted ones.

III.1. INTRODUCTION

Intended to reach a "good ecological status" of all water bodies by 2015, Europe's Water Framework Directive (WFD) has prompted a large amount of works which yielded characterizations of either reference physical-chemical environments and biological communities in continental Europe, as well as practical tools (e.g. biological indices) to evaluate water quality (Borja, 2005). Overseas regions of Europe occur in various biogeographic areas of the World (Atlantic, Caribbean, Pacific, Indian Oceans). These regions have the same water policy objectives as the continental ones, but they were overlooked during recent developments of bioassessment tools that fulfill the WFD guidelines. Differences in bioclimatic, biogeographic and geomorphological conditions, as well as differences in anthropogenic pressure preclude the transposition of typological schemes and bioassessment tools developed in continental Europe to overseas regions. For instance, biological traits (life history patterns, body size, etc.), species richness and numerical dominance do not compare among biogeographic regions. Last but not least, the development of bioassessment methods in many overseas regions suffer from a lack of taxonomic knowledge, so that ecologists are faced with minimal background on the distribution patterns of aquatic species. For instance, very little is known about macroinvertebrate taxonomy and distribution in headwater streams of French Guiana, East-Amazonia.

French Guiana (FG) is an overseas region of France located on the north-eastern coast of South America. About 96% of its surface area (83,534 km2) is covered by a remarkably species-rich equatorial forest (Bongers et al., 2001). The Guianese primary forest remains one of the least impacted of the World, however, gold mining and timber have strong localized impacts upon river ecosystems. Specifically, the annual gold output in the area is 60 times higher than 25 years ago (Hammond, 2007). After the prospection of large rivers, gold industries are now focusing on smaller inland streams (Cleary, 1990; Hammond, 2007). Small streams (from headwaters to rivers with depth< 1m and width < 10m) represent 70-80% of all running waters in FG. Most small streams are located in forested areas and exhibit high ecological quality; some if not most of them have never been impacted by any human activity. In light of recent economic development, there is a pressing need to identify reference (undisturbed) conditions that will then allow environmental managers to estimate the degree to which human activities have altered stream ecosystems.

While providing new basic information on freshwater diversity and its environmental drivers in eastern Amazonia, this study takes a step towards the implementation of the WFD in

French Guiana (one of France's 11 inhabited overseas regions) by bringing out the first classification of FG streams. Routine surveys conducted by local consultancies revealed changes in river communities in relation to local anthropogenic pressure (Vigouroux et al., 2005). However, because FG is mostly covered by dense (inhabited) rainforest deprived of road networks, the local to regional distribution patterns of macroinvertebrates are fundamentally unknown, especially in the remote, headwater streams. By using ordination and classification of 65 sampling sites distributed throughout FG, we tested the following predictions: (i) geomorphology determines ecological sub-regions that have typical macroinvertebrate assemblages and species richness, and (ii) invertebrate diversity broadly declines as anthropogenic pressures increases. Environmental explanatory variables were used to interpret invertebrate diversity and distribution, and the resulting schemes were discussed in the context of water policy.

III.2. MATERIAL AND METHODS

III.2.1. Study Area

This study was conducted in French Guiana, from September 2011 to December 2012. The climate is wet tropical with 3,000 - 3,400 mm of annual precipitation mainly distributed over 280 days. There is less rainfall (dry season) between September and December and another shorter and more irregular dry period in March. The average monthly maximum temperature is 33.5°C (32.1-35.8°C), and the average monthly minimum is 20.3°C (19.7-21°C). French Guiana's streams flow into seven large river watersheds (Maroni, Kourou, Mana, Sinnamary, Comté, Approuagues, and Oyapock rivers). It is worth noting that "small streams" (water depth < 1m; stream width < 10m) represent ca. 80 000 km in total length, i.e. 70-80% of all running waters in the region.

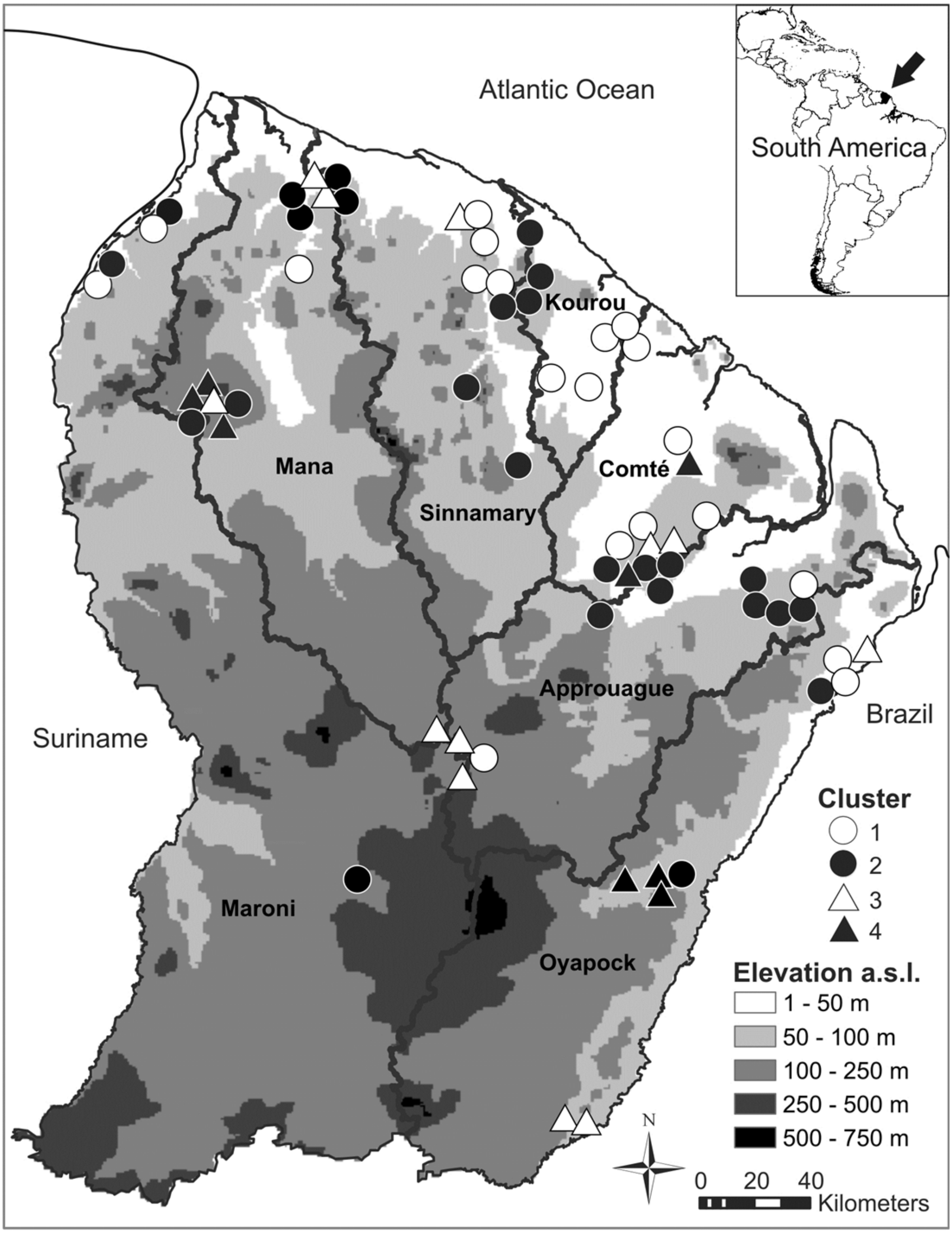

Figure 27: Distribution of the 65 sampling sites in French Guiana. Different markers are used to assign sites to clusters 1-4 depicted in Figure 28.

III.2.2. Sampling sites and environmental variables

We sampled 65 sites belonging to different watersheds distributed throughout the country (Figure 27). The sampling effort was inevitably higher in the northern range, due to the limited access to the south. The complex logistics needed to obtain samples from this remote range limited the number of sites. We nevertheless managed to sample southern sites from the main river basins (Figure 27). All sites were sampled during the dry season (September-December) in 2011 and 2012. Impacted sites were subjected to two major disturbance types: gold mining (n= 11; either legal or illegal), and land-use impacts (n= 14; logging for wood products and timber, runoff from small cultivations and/or urban areas). Reference sites (n= 40) were defined as sites not subjected to anthropogenic impacts.

Table VIII: Main physical-chemical characteristics of the 65 samplings sites in French Guiana.

	Mean ± SE	Minimum–maximum
Topology		
Distance from source (km)	3.52 ± 3.49	0.5–16
Elevation (m.a.s.l)	171.6 ± 78.04	32.46–234.64
Slope (per mil)	3.66 ± 2.79	0.34–11.58
Mean stream width (m)	4.06 ± 2.27	1.04–13.56
Mean stream depth (cm)	27.4 ± 11.81	5.25–55.06
Substrate composition		
% Silt	15.54 ± 17.92	0–90
% Sand	45.21 ± 31.55	0–100
% Gravel	20.37 ± 20.79	0–85
% Coarse substratum	18.93 ± 24.95	0–85
% Woody debris	12.94 ± 10.4	0–41.6
% Macrophytes	10.56 ± 16.46	0–70
% Litter	21.92 ± 19.89	0–85
% Roots on the bank	12 ± 10.53	0–50
Water chemistry		
pH	5.73 ± 0.79	4.04–7.65
Temperature (°C)	24.96 ± 1.53	19.9–30.7
Conductivity (μs cm^{-1})	32.62 ± 24.69	12–135
Dissolved oxygen (mg l^{-1})	6.37 ± 1.45	3.7–7.9
Turbidity (NTU)	12.37 ± 44.87	0.51–329
Total suspended matter (mg l^{-1})	16.3 ± 70.07	0–566
Dissolved organic carbon (mg l^{-1})	18.15 ± 14.38	0–78
Nitrate (μg l^{-1})	0.29 ± 0.15	0–0.83
Total phosphorus (μg l^{-1})	0.04 ± 0.04	0–0.172

All sampling sites were characterized using topological, morphological, water chemistry, and habitats variables (Table 1). For each site, a Geographic Information System (GIS, ESRI ArcGis 10) was used to obtain elevation above sea level (m a.s.l.), distance from the source (km), and slope (per mil). These variables were chosen because they characterize the location of sites within the upstream-downstream river continuum. Water samples for chemical analyses were taken at each site between 10:00 am and 2:00 pm (to minimize potential diurnal variation in the data), and immediately frozen. Chemical analyses were carried out at Hydreco Laboratory (Petit-Saut, French Guiana) following standardized methods (AFNOR 2000, 2005a, 2005b). Chemical variables measured in the laboratory were: turbidity (NTU), Total Suspended Matter (mg.L-1), NO3 (µg•L-1), Total Phosphorus (µg•L-1)), and Dissolved Organic Carbon (mg L-1). Four variables were directly measured in the field using probes: % oxygen (WTW 3205®), turbidity (EUTECH®), pH (WTW 3110®) and conductivity (WTW 3110®). Water temperature (°C) was the mean of values given by all above-mentioned probes.

The length of a site was defined as 10 times its width, and transects were established each 5 meter along this length, for subsequent habitat measures. Water depth (m) and the percentage composition of organic and mineral substrate types were determined on a 1m2 area every meter along each transect. Mean water depth at a site was the mean of all point measurements. Stream width (m) was the mean value of all transects. The substrate types included: % litter, % submerged roots on the banks, % macrophytes, % woody debris, % silt, % sand (particle size <2mm), % gravel (2-25mm), % coarse substratum (> 25mm). Coarse substrates being scarce in FG streams, pebbles, boulders, and rocky outcrops were grouped in a single category.

III.2.3. Macroinvertebrates Sampling

Twelve sample units were taken at each site, i.e. 8 samples in organic substrates (roots, macrophytes, aquatic plants, litter, bryophytes) and 4 samples in mineral substrates (pebbles, gravels, sand), thus representing the average distribution of these substrate types in FG streams. Sample units in organic substrates consisted in intensive sweeping of a hand net (frame size= 46 x 23 cm; mesh size = 500µm) during 1 minute over a 0.46x1.5m area (net width x 1.5m). Sample units in mineral substrates were obtained by dragging a 5cm-layer of sediment with the same net, over a 0.46x1.5m area. Prior to dragging, coarse particulates (pebbles) were brushed in front of the net, and then removed. The samples were preserved in the field in 4% formalin (final concentration). Invertebrates were sorted in the laboratory and preserved in 70% ethanol. They were mostly identified to family and enumerated (list of taxa and mean numbers of individuals per m2 in electronic supplementary material).

III.2.4. Data analysis

To sort the 65 sampling sites according to the invertebrate communities, we used the Self-Organizing Map algorithm (SOM Toolbox version 2 for Matlab, see Vesanto et al. (1999) for practical instructions). The strengths of the SOM in comparison with conventional multivariate analyses were discussed in Giraudel and Lek (2001). Briefly, combining ordination and gradient analysis functions, the SOM is convenient to visualize high-dimensional data in a readily interpretable manner without prior transformation. Here, it is worth noting that conventional (multivariate) ordination and classification techniques were inefficient at revealing patterns of community organization, certainly because we had to analyse organism counts with skewed distributions (due to many zero values). The SOM algorithm is an unsupervised learning procedure that transforms multi-dimensional input data into a two-dimensional map subject to a topological (neighbourhood preserving) constraint (Kohonen, 2001). The SOM thus plots the similarities of the data by grouping similar data items together onto a 2D-space (visualized as a grid) using an iterative learning process (Park et al. 2003). The SOM algorithm is specifically relevant for analyzing sets of variables that vary and co-vary in non-linear fashions, and/or that have skewed distributions. Additionally, the SOM algorithm averages the input dataset using weight vectors through the learning process and thus removes noise. A full description of the modeling procedure employed here (training, map size selection, number of iterations, map quality measurements) was detailed in Céréghino & Park (2009).

The structure of the SOM for this analysis consisted of two layers of neurons connected by weights (or connection intensities): the input layer was composed of 86 neurons (one per invertebrate taxon) connected to the 65 sampling sites, and the output layer was composed of 42 neurons visualized as hexagonal cells organized on an array with 7 rows and 6 columns. The number of 42 output neurons was retained after testing quantization and topographic errors (see Céréghino & Park, 2009). At the end of the training, each site is set in a hexagon of the SOM map. Sites appearing distant in the modeling space (according to invertebrate data used during the training) represent expected biological differences for real environmental characteristics.

Ward's algorithm was applied to cluster the trained map (Ultsch, 1993). The SOM units (hexagons) were divided into clusters according to the weight vectors of the neurons, and clusters were justified according to the lowest Davis Bouldin Index, i.e. for a solution with low variance within clusters and high variance between clusters (Negnevitsky, 2002).

In order to analyze the contribution of each invertebrate taxon to cluster structures of the trained SOM, each input variable calculated during the training process was visualized in each neuron (hexagon) of the trained SOM in grey scale. This visualization method directly describes the discriminatory powers of input variables (here invertebrates) in mapping (Kohonen, 2001), while allowing to bring out invertebrate distribution patterns. To investigate relationships between physical-chemical and biological variables, we introduced the 22 physical-chemical variables into the SOM previously trained with the abundance data for the 86 invertebrate taxa (see Céréghino & Park, 2009). During the training, we used a mask function to give a null weight to the 22 physical-chemical variables, whereas biological variables were given a weight of 1 so that the ordination process was based on the 86 invertebrate taxa only (Compin & Céréghino, 2007). Setting mask value to zero for a given component removes the effect of that component on organization (Sirola et al., 2004).

Model structures (clusters of sites) were visualized using GIS. Significant differences in taxa richness, evenness (Simpson index) and entropy (Shannon-Weaver index) among SOM clusters were tested using Kruskal-Wallis tests.

III.3. RESULTS

After training the SOM with the invertebrate densities at 65 sites, the sites were classified into four subsets (clusters 1 to 4) according to the quantitative structure of their macroinvertebrate communities (Figure 28). Clusters were plotted on a geographical map of FG in order to ease interpretations (Figure 27). Sites in clusters 1-2 and 3-4 corresponded to two major geographic areas of FG, i.e., coastal areas and inland forests, respectively. Within coastal ranges, sites subjected to logging (TETE, KAP1, KAP2, KAP 4 and KAP 5) and gold mining (KORO, ROSE, BORD, GREN, BOIB) were grouped in cluster 2 in the left-bottom area of the map. All sites subjected to agricultural or urban runoff (LUCI, REL1, REL2, DACH, PLAM, SM, APA, BAST, HUMU) belonged to coastal areas, but did not show clear grouping. Within inland forest ranges, sites submitted to gold mining (QUAD, CLAN, CHS2, CHAF) were grouped in cluster 4 in the left-top area of the map. Therefore, based on site status (reference vs impacted) each major geographic area was further separated into two sub-groups of sites according to a gradient of anthropogenic impact ranging from low (right area of the SOM, clusters 1 and 3) to high (left area, clusters 2 and 4).

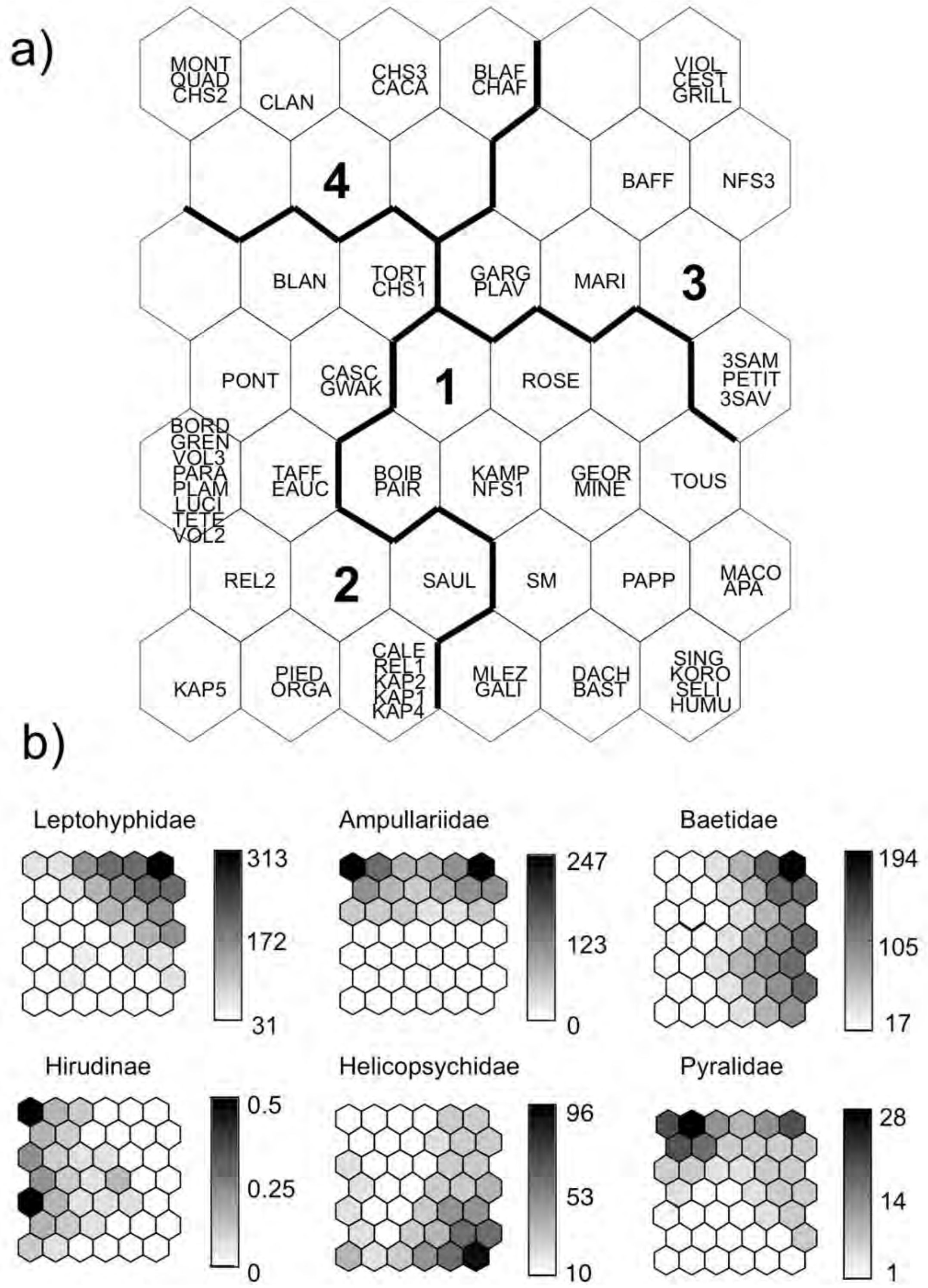

Figure 28:(a) Distribution and clustering of the 65 sites on the self-organizing map (SOM) according to the abundance of 86 macroinvertebrate taxa. Codes within each hexagon (e.g., MONT, QUAD) correspond to sites. Clusters 1 to 4 were derived from Ward's algorithm. (b) Gradient analysis of density (number of individuals per m^2) for a few selected taxa on the trained SOM represented by a shaded scale (dark: high density, light: low density). Each small map representing taxa that follow similar patterns can be compared to the map representing the distribution of sites in (a), thus showing the distribution patterns of the various taxa (in shades of gray) within each sub-area of the SOM.

Only 9 taxa out of 86 (e.g., Noteridae, Sialidae, Lestidae) occurred in one specific cluster of sites (see electronic supplementary material). When the distribution of each taxa was visualized on the trained SOM using a shading scale (examples in Figure 28b), Baetidae and Caenidae (Ephemeroptera), Notonectidae (Heteroptera), Limoniidae (Diptera) and Polycentropodidae (Trichoptera) were characteristic of unimpacted sites, whatever the geographic area (clusters 1 and 3). Hirudinae, Oligochaeta and Nematodes were frequent in impacted sites (clusters 2 and 4). Higher densities for these taxa therefore indicated anthropogenic impacts, rather than regional differences in stream habitat conditions. Sites in forest ranges (clusters 3-4) showed higher densities (and occurrences) for invertebrate families belonging to the Mollusca (Ampullariidae, Hydrobiidae and Thiariidae) (see examples in Figure 28b). Such taxa therefore had strong influence upon the classification. Cluster 3 (forest, reference sites) showed higher densities for Leptohyphidae and Leptophlebiidae (Ephemeroptera), Megapodagrionidae and Calopterygidae (Odonata), Ceratopogonidae, Empididae, Culicidae and Simuliidae (Diptera), Elmidae (Coleoptera) and Planaria. Cluster 4 was characterized by higher densities for Ephemeroptera (Euthyplociidae), Plecoptera (Perlidae), Dryopidae (Coleoptera), Lepidoptera (Pyralidae), Psychodidae (Diptera) and Odonata (Plastytiscidae). Sites in coastal ranges (clusters 1 and 2) had a lower number of typical taxa, namely Coryphoridae (Ephemeroptera), Euryrhinchidae and Helicopsychiidae (Trichoptera). Sites in the cluster 1 were characterized by higher densities of Helicopsychidae, Glossosomatidae and Odontoceridae, (Trichoptera), Corethrellidae (Diptera), Coryphoridae and Polymitarcyidae (Ephemeroptera). Sites in cluster 2 were characterized by low numbers of taxa and individuals.

Finally, the comparison of community diversity indices between clusters only showed significant differences for taxonomic richness: the number of invertebrate taxa was significantly lower in cluster 2 than in cluster 1 ($p= 0.0015$) and cluster 3 ($p= 0.015$, Figure 30).

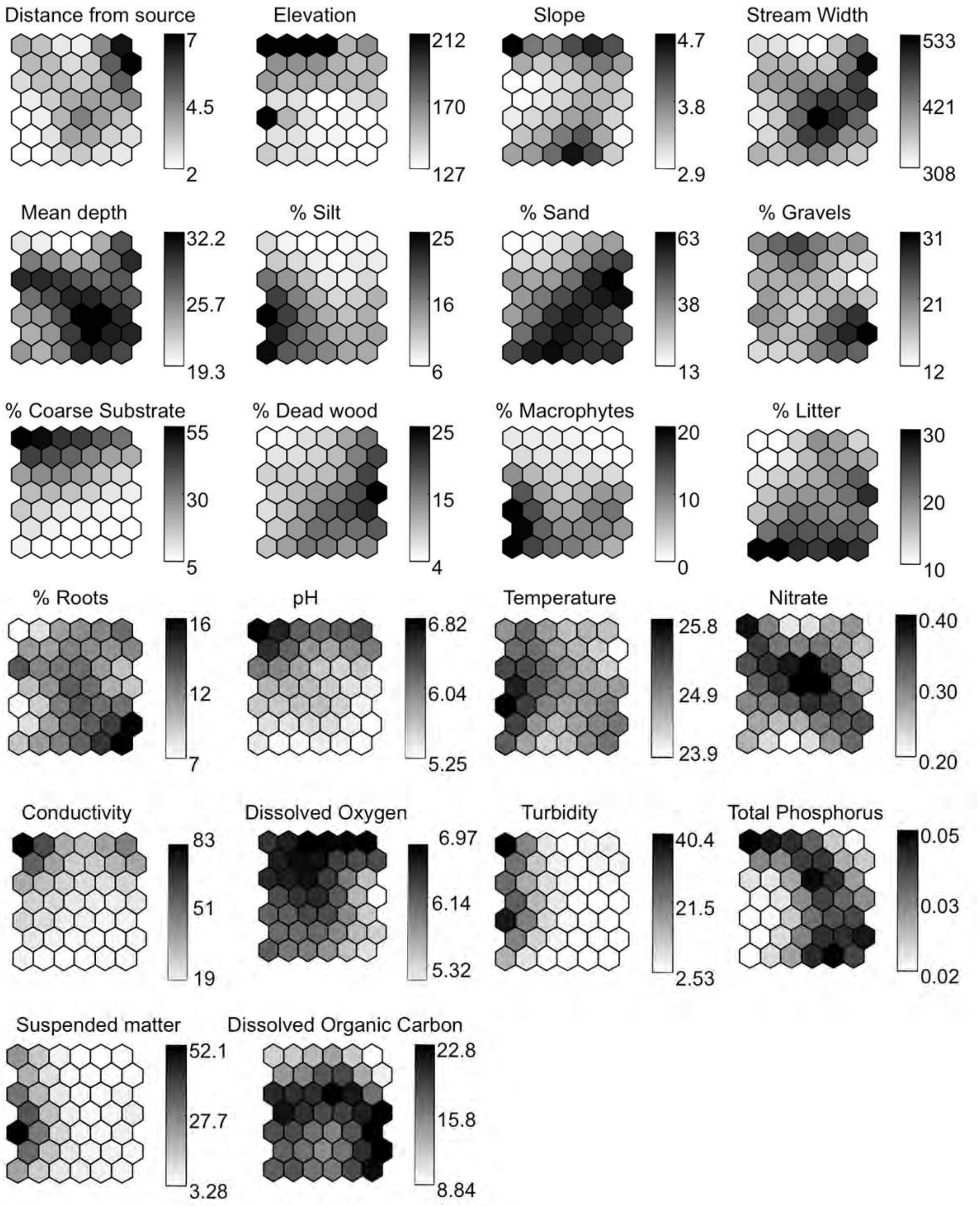

Figure 29: Gradients of selected environmental variables on the SOM previously trained with macroinvertebrate data. The mean value for each variable was calculated in each output neuron of the SOM. Dark represents a high value, while light is a low value. See Table VII1 for units.

When environmental variables were introduced into the SOM previously trained with abundance data for 86 invertebrate taxa at the 65 sites (Figure 29), sites in clusters 1-2 were characterized by finer substrates (sand and silt) as well as higher %macrophytes, %litter and %dead wood. Sites in clusters 3-4 had higher values for elevation, conductivity, pH and %coarse mineral substrates. Specifically, physical-chemical variables that indicate human impacts (higher values for turbidity and suspended matters from right to left areas of the map) confirmed the gradient of disturbance within each sub-region, from cluster 1 to 2, and 3 to 4.

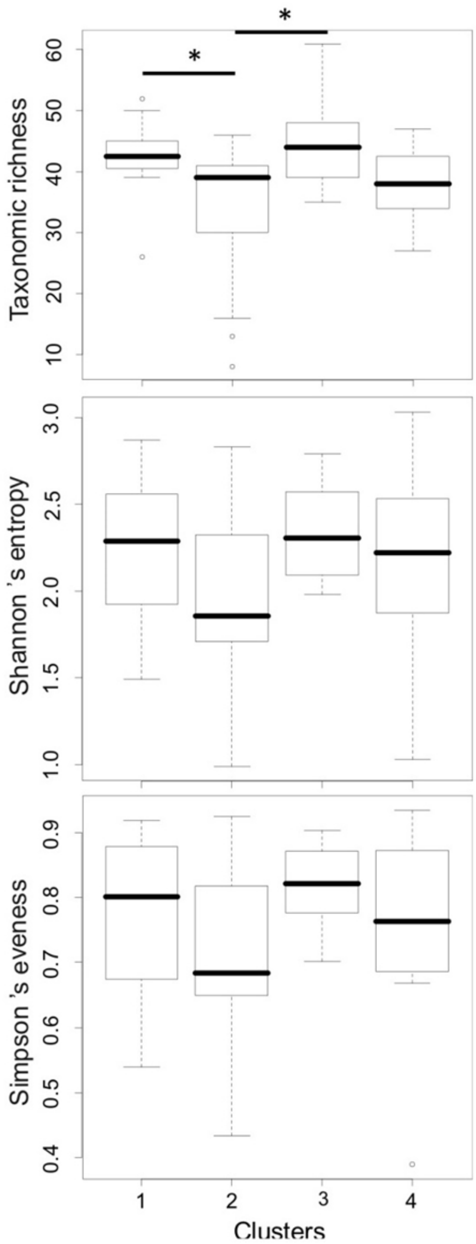

*Figure 30: Boxplots of diversity metrics distributions (taxonomic richness, Shannon's Entropy, Simpson's evenness), in the four clusters derived from the SOM clustering. Significant differences between groups or clusters were tested with Kruskal-Wallis tests; *= significant differences at P<0.05.*

III.4. DISCUSSION

This study provides new information on the environmental determinants of freshwater invertebrates diversity and taxa distribution in eastern Amazonia, while proposing the very first biological typology of FG streams. Previously, Chandesris (2005) suggested an abiotic typology of FG watersheds by delineating hydro-ecoregions based on geomorphological, hydrological, and climate data. Delineated according to macroinvertebrate communities, clusters 1 and 2 in our study match the "coastal alluvial plain" characterized by recent sediment and low elevations, while clusters 3 and 4 correspond to the inland "Guiana Shield" characterized by an eroded rocky substrate, a variability of elevations and large stream systems under a dense forest coverage. These results also indicate that in the streams of FG, the structure of macroinvertebrate communities changes along a longitudinal gradient, from inland headwaters to the coastal rivers. First of all, sites in cluster 3-4 had coarse bed paving substrates, and hosted richer and more diverse invertebrate communities than the sandy sites of cluster 1-2. Downstream changes in tropical stream invertebrate communities were previously related to changes in ecological processes along gradients of elevation (i.e. from up- to downstream areas), notably changes in leaf litter inputs and algal production (Sites et al., 2003). In FG however, there is no such clear gradient of elevation above sea level. Inland forest ranges must be seen as a dense "sea of hills" forming higher elevation islands within a low-elevation matrix. Hence, on a local scale, steep slopes alternate with long flat plateaus. Because river competence determines substrate size and, subsequently, invertebrate diversity (Buss et al., 2004; Arrington & Winemiller, 2006; Salman et al., 2013), some inland sites close to the headwater sources can group together with coastal sites within the framework of a biological typology (e.g. sites KAMP CALE and NFS1).

There was also a clear difference between clusters of sites in terms of pH. Coastal sites correspond to acidic waters, locally called "black waters" because of their darker colour. pH values in headstream waters were neutral. Only a few invertebrate families were strictly characteristic of neutral waters however, suggesting that most taxa have broad pH tolerance. Those taxa specific of neutral streams belong to Mollusca and Ephemeroptera. The formation of the shell of freshwater mollusca notably requires neutral to basic pH (Merritt & Cummins, 1996). The sensitivity of Ephemeroptera to acidification has been previously demonstrated in temperate rivers (Dangles and al., 2004; Petrin et al., 2007) but not in tropical areas yet. Insect families like Euryrhinchhidae, Corydalidae, Odontoceridae Helicopsychidae and the Ephemeroptera Polymitarcidae are mainly found at low pH in FG streams, meaning that they tolerate acidity. However, because of their life style, these taxa also have a strict preference for a given substrate type (Merritt & Cummins, 1996). Odontoceridae and Helicopsychidae caddisflies require sand to build their larval case, and the Polymitarcidae is a specialist burrower in silt and sandy substrates. Coryalidae are mainly observed under dead wood. Such striking, selected examples support the hypothesis that in naturally acid stream, community composition does not only depend on water acididty but also on substrate size.

Ordination and cluster analyses are frequently used in the exploratory phase of typologies. All sites were included in our SOM, regardless of a priori consideration of disturbance. By doing so, we expected that geographically adjacent sites appearing distant in modelling space (according to macroinvertebrate communities) would represent differences among sites in biological quality. Sites subjected to anthropogenic disturbance grouped in specific clusters within large hydro-ecoregions, suggesting that disturbance have an effect on freshwater diversity but did not override geomorphological controls of the distribution of macroinvertebrates in FG streams. Gradients of disturbance were apparent both within coastal plains and forest ranges, revealing ecological impacts of gold mining and logging . Previous studies demonstrated that bank erosion due to these activities increase sediment upload, to the detriment of freshwater diversity (Cleary, 1990; Mol & Ouboter, 2004; Hammond, 2007). In addition, gold miners extract and crush coarse mineral substrate, further homogenising river beds and generating high turbidity that decrease light penetration into the water to the detriment of epilithic algae, an important base of the food chains for invertebrate (Sloane-Richey et al., 1981; Graham, 1990). Interestingly, a few impacted sites located in the inland forest, namely TAFF, BORD, TORT and GREN, were assigned to cluster 2 (impacted sites, coastal area) instead of cluster 4 as we could have expected. These sites are typically subjected to small-scale illegal mining. Illegal settlements are increasingly located in remote areas, and are not cleared by gold miners in order to remain invisible to aerial and satellite surveillance (Hammond, 2007; Coppel et al.; 2008). While former illegal gold mining concentrated on downstream river reaches, there is little information on the impact of this new trend of gold mining activities (Mol & Ouboter, 2004; Mendiola, 2008; Yule et al., 2010; Brosse et al., 2011). Our analyses therefore suggest that gold-mining in headwater, forested streams may have the harsher impact upon freshwater diversity because it is likely to generate strong longitudinal discontinuities, making upstream communities resembling the most downstream impacted ones.

All sites subjected to less severe disturbance (runoff from small cultivations and/or urban areas) were located in the coastal alluvial plain (clusters 1 and 2), which concentrates 80 % of FG's human population. The distribution of these sites did not follow a clear gradient of disturbance within the corresponding hydro-ecoregion, suggesting that small-scale increases of dissolved organic carbon and/or nitrogen and/or phosphorus concentrations that typically match agricultural and/or urban activities were not large enough to generate significant deviation from predictable communities. Given that nutrient inputs are limited to diffuse runoff from small cultivations and/or sparse habitations, we assume that, in the absence of gold-mining and timber, the proportions of fine mineral substrate and organic substrates (submerged roots, macrophytes, dead wood, litter) play a key role in determining invertebrate diversity at these coastal sites.

Whilst Europe's WFD provided compelling reasons for developing river typologies, reference schemes, and pressure-impact models in member States, the lack of published study for overseas regions first reflects minimal knowledge of the distribution patterns of aquatic species in "neotropical Europe", especially in remote headwaters (but see recent works by Bernadet et al. 2013 and Touron-Poncet et al. 2013 in the Carribean). Our analysis revealed that invertebrate communities show qualitative and quantitative spatial patterns, but also change in terms of biological traits in relation to natural conditions and anthropogenic disturbance (e.g., increasing mollusk diversity in forest sites, insects at reference sites, annelids at impacted sites). Hence, our typology will prove useful in defining impacted and least impacted river reaches for the upcoming development of a WFD-compliant biological index for FG (Mondy et al., 2012). Our study however shows that human pressure have an impact on FG streams but no clear gradient of disturbance was observed, i.e. our impacted sites were subjected either to negligible (diffuse runoff) or harsh disturbance (gold-mining, logging). In other words, intermediate disturbance is clearly lacking in FG. Assuming that modern biological indices must be scaled against a gradient of water quality corresponding to different levels of impairment (typically representing "high", "good", "moderate", "poor" and "bad" ecological quality), we may anticipate difficulties in defining intermediate quality classes between "bad" and "good" quality. Hence, further analyses of physical-chemical environments would be needed to identify tipping points between natural and disturbed states.

Chapitre IV. Un indice multimétrique basé sur les invertébrés pour la mise œuvre de la directive cadre européenne sur l’eau en Guyane Française

Ce travail a fait l'bbjet d'une publication dans la revue scientifique River Research and Applications.

Résumé

Bien que distantes du continent, les Départements d'Outre-Mer (DOM) font partie intégrante de l'Union européenne et sont soumis aux mêmes objectifs en termes de politiques environnementales. Les DOM ont cependant été négligés lors des premiers développements de bio-indicateurs qui répondent aux exigences de la Directive Cadre sur l'Eau. Nous proposons un indice multimétrique DCE-compatible, applicable aux petites masses d'eau de Guyane. L'étape essentielle qui est présentée ici concerne l'étude des liens entre pressions anthropiques et état écologique. Ces relations pressions/état conditionnent le choix des métriques constitutives du futur indice. Les métriques retenues devaient en outre suivre les prescriptions par la DCE en prenant en compte : i/ des conditions naturelles pour exprimer résultat de l'évaluation d'un site en terme « d'écart à la référence » pour le type de cours d'eau considéré, ii/ de la diversité, iii/ de l'abondance et iiii/ de la sensibilité des taxons.

A partir d'un jeu de donnée de 95 sites, nous avons conçu l'Indice Biologique Macroinvertébrés de Guyane (IBMG) pour évaluer la qualité écologique des petits cours d'eau. Parmi les 103 métriques biologiques mesurées à partir de données stationnelles, nous avons sélectionné les paramètres présentant le meilleur compromis entre : haute efficacité de discrimination, faible spécificité, faible redondance et stabilité dans les conditions de référence. L'IBMG est composé de deux métriques basées sur la richesse taxonomique, deux sur l'abondance, une sur un indice de diversité et dernière liée à un trait fonctionnel. Chacune des métriques a été pondérée par son efficacité discrimination au sein de l'indice. L'utilisation d'un jeu de données test a permis de démontrer que l'IBMG est sensible à la gamme des perturbations rencontrées en Guyane. Cependant, une métrique basée sur la sensibilité aux pollutions n'a pas pu être intégrée à l'indice actuel, en lien avec le manque de résolution taxonomique et de connaissances sur l'écologie de la faune néotropicale. Enfin, la comparaison de l'IBMG avec les autres indices développés dans la région neotropicale a révélée que, pour plusieurs raisons, les indices multimétriques développés dans les néotropiques tendent à être efficaces dans le contexte régional ou ils ont été construits, mais ils sont certainement moins robustes à plus large échelle biogéographique.

A multimetric macroinvertebrate index for the implementation of the European Water Framework Directive in French Guiana, East-Amazonia.

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Short title: A multimetric macroinvertebrate index for East-Amazonian streams

Abstract

Neotropical, overseas regions of Europe are subjected to same water policy objectives as the continental ones, but were overlooked during recent developments of bioindicators that fulfill the Water Framework Directive guidelines. We designed a macroinvertebrate-based multimetric index (IBMG) to assess ecological health in remote headwater-small streams of French Guiana, Europe's only overseas region of continental South-America. Invertebrates were sampled at 95 sites including reference and impacted river reaches, following a standardized protocol. Among the 102 biological metrics calculated from site-specific data, we selected metrics exhibiting the best trade-off between high discrimination efficiency, low specificity, low redundancy, and high stability under reference conditions. The IBMG is composed of two taxonomic richness-based metrics, two abundance-based metrics, one trait-related metric and a diversity index (Shannon's entropy). Each metric was weighted by its discrimination efficiency. Using a test data set, we found that the IBMG was sensitive to the range of disturbances in FG. Finally, comparing the IBMG with other indices developed in other neotropical countries reveals that, for several reasons, multimetric indices developed in the neotropics may perform well in the context of the datasets used to generate them, but would certainly fail to be robust when used elsewhere.

Keywords: anthropogenic perturbation; biological diversity; biomonitoring; neotropical rivers; reference conditions

IV.1. INTRODUCTION

Intended to reach a good ecological status for all surface waters by 2015, Europe's Water Framework Directive (European Council, 2000) has set up unified guidelines for the design and implementation of biological assessment tools in member states. After a decade of research intended to classify surface waters into biogeographic and geomorphological types, to agree upon reference (pristine) conditions for each type, and to design biological indices that measure ecological health in terms of similarity to a reference state (Bailey et al., 1998), recent examples of WFD-compliant bioassessment tools in continental Europe can be found, for instance, in Gabriels et al., 2010 (Belgium); Kelly et al., 2012 (Ireland); Mondy et al., 2012 (France). European Overseas Countries and Territories (OCTs) and Outermost Regions (OMRs) occur in most biogeographic areas of the World, from polar to tropical ranges. Although inclusion in EU policies may vary among OCTs and OMRs, most of these territories are subjected to WFD's objectives. However, these regions were overlooked during the development of methods that fulfil WFD's requirements, at the point that most if not all of them still lack WFD-compliant tools. This is particularly true of the French overseas departments, e.g., Martinique and Guadeloupe in the Carribean (but see Touron-Poncet et al., 2014 for recent updates), Reunion Island in the Indian Ocean, French Guiana in the Eastern Amazon. Differences in climatic, biogeographic and geomorphological conditions, as well as differences in anthropogenic pressure preclude the transposition of bioassessment tools developed in continental Europe to overseas regions. Clearly, biological traits (habitat preferences, sensitivity to pollution, etc.), species richness and numerical dominance do not compare among biogeographic regions. Finally, applied river research in many overseas regions suffer from a lack of taxonomic knowledge, so that ecologists are faced with minimal background on the distribution patterns of aquatic species. It is worth noting however that most OCTs and OMRs are small islands, so their freshwaters are expected to host depauperate faunas compared to their continental counterpart (Bass, 2003; Boulton et al., 2008). Nevertheless, the taxonomic issue is more pressing in species rich continental areas of the tropics. For instance, very little is known about macroinvertebrate taxonomy and distribution in remote headwater streams of French Guiana, East-Amazonia. Importantly, biological monitoring methods are generally less expensive than chemical methods, and this could be important in South-American countries.

In French Guiana (FG), like in other South-American countries, small streams are located in forested areas and exhibit high ecological quality; some if not most of them have never been impacted by any human activity. Although FG's primary forest remains one of the least impacted of the world, gold mining and timber for wood products have strong impacts upon river ecosystems. A detailed description of the physical-chemical impacts of logging and gold-mining in streams of FG was given in Dedieu et al. (2014). In particular, the annual gold output of FG is 60 times higher than 25 years ago (Hammond et al., 2007) and after the exploitation of large rivers, gold mining is now clearly shifting towards smaller inland streams (Hammond et al., 2007; Brosse et al., 2011). Unfortunately, no biological indices or any form of biological indicators are available yet for small streams of FG. Given recent economic development, there is, therefore, a pressing need to implement WFD-compliant biological assessment tools in FG, so that future water quality assessments and objectives can be established in light of current ecosystem health and economic plans. As part of the EU water policy, multimetric indices based on Biological Quality Elements (BQEs, e.g., diatoms, benthic macroinvertebrates, fish, macrophytes) became the standard to evaluate ecosystem health. Multimetric indices assemble and weight different types of metrics (e.g. taxonomic richness, abundance, functional traits) that effectively respond to environmental heterogeneity and impairment, and therefore provide more accurate assessment of ecosystem health than single metrics (Barbour et al., 1999; Hering et al., 2006, 2010). In an earlier study, Chandesris and Wasson (2005) have delineated hydro-ecoregions (HERs) of FG based on geomorphological, hydrological and climatic data. In addition, Dedieu et al. (2015) have classified small streams of French Guiana, based on extensive sampling of macroinvertebrate (the most widely used BQEs to date) and physical-chemical data. Importantly, these works allowed us to identify stream types and reference conditions for FG streams, thus providing background for the development of WFD-compliant biological indices.

In the present study, we take a step towards the implementation of the WFD in Europe's only overseas department of continental South-America, by proposing a multimetric index based on river macroinvertebrate diversity. In order to address instructions for consistency in methods imposed by national environmental agencies, we mostly followed the methodology established in metropolitan France by Mondy et al. (2012), with adaptations inherent to biogeographic differences in community diversity, fundamental knowledge of species/population ecology, and anthropogenic impacts. We assessed the efficiency of our new multimetric index on a test data set. Finally, we discuss our results in comparison to other multimetric indices recently developed in the neotropics with the broader aim to highlight gaps in knowledge about (bio)geographic differences in ecosystem structure and functioning, as well as human activities that influence them.

IV.2. MATERIAL AND METHODS

IV.2.1. Study area

This study was conducted in French Guiana (surface area = 83,534 km^2), East Amazonia, from September 2011 to December 2012. The climate is tropical moist with 3,000 – 3,400 mm of yearly precipitation mainly distributed over 280 days. There is a major drop in rainfall (dry season) between September and December and another shorter and more irregular dry period in March. The maximum monthly temperature averages 33.5° C (32.1-35.8°C), and the monthly minimum averages 20.3°C (19.7-21°C). French Guiana's stream systems are organized around seven large rivers (Maroni, Kourou, Mana, Sinnamary, Comté, Approuagues, and Oyapock); however, "small streams" (from headwaters to rivers with depth< 1m and width < 10m) represent ca. 80 000 km in total length, i.e. 70-80% of all running waters in the region.

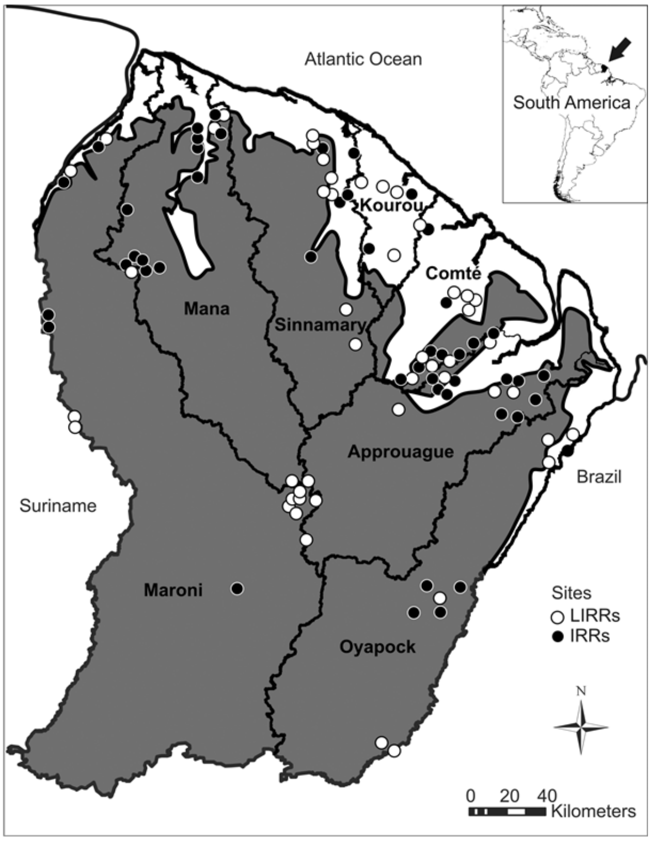

Figure 31: Location of the sampling sites in French Guiana. The two different shades illustrate the sub-regions (white: costal alluvial plain, grey: Guiana Shield) that form stream types according to hydro-ecoregions and macroinvertebrate assemblages (see text). Markers indicate the status of the sites: Least Impacted River Reaches (LIRRs) and Impacted River Reaches (IRRs).

IV.2.2. Field sampling

We sampled 95 sites, distributed over 76 small streams belonging to FG's main drainage basins (Figure 31). It should be noted that the sampling effort was higher in the Northern part of FG, due to the difficulty to access southern FG. In this specific area covered by dense rainforest and without road networks, complex logistics limited our ability to sample a larger number of sites. We however managed to collect some samples from the main southern river basins. All sites were sampled during the dry season in 2011 and 2012 (September-December). Indeed, most remote sites were not accessible during the rainy season. In addition, human perturbation is detected less efficiently during high flows because of dilution effect. Additionally, we sampled 26 sites, including 14 sites subjected to human impacts (e.g., logging, gold mining). These new data sets were used as 'test' data sets.

Physical variables were chosen to describe the heterogeneity of the river bed substrate at each site. They were recorded directly in the field and accounted for the percentage composition of organic and mineral substrate types, using the standardized protocol by Souchon et al. (2000). These variables included: % leaf litter % submerged roots on the banks, % submerged vegetation, mostly macrophytes, % woody debris, % silt, % sand (< 2mm), % gravel (2-25mm), % coarse substratum (> 25mm). Coarse substrates being scarce in French Guiana, this category of mineral substrate included pebbles, boulders, and/or rocky outcrops.

Water samples for chemical analyses were taken at each site and immediately frozen. Chemical analyses were carried out at Hydreco Laboratory (Petit-Saut, French Guiana) following standardized methods (AFNOR 2000, 2005a, 2005b). Chemical variables measured in the laboratory were: total suspended matter ($mg.L^{-1}$), nitrate ($\mu g.L^{-1}$), total phosphorus ($\mu g.L^{-1}$) and dissolved organic carbon ($mg.L^{-1}$). Four variables were directly measured in the field using probes: dissolved oxygen ($mg.L^{-1}$) (WTW 3205®), turbidity (NTU) (EUTECH®), pH (WTW 3110®) and conductivity ($\mu S.cm^{-1}$) (WTW 3110®). Water temperature (°C) was the mean of values given by all above-mentioned probes.

To collect the benthic macroinvertebrates, twelve sample units were taken at each site, i.e., eight samples in organic substrates (group A samples, e.g. submerged vegetation, leaf litter) and four samples in mineral substrates (group B samples, e.g. sand, gravel). This distribution of samples was based on the above-mentioned description of the various organic and mineral substrates in our small streams. Sample units in organic substrates consisted in intensive sweeping of a hand net (frame size= 46 x 23 cm; mesh size = 500µm) during 1 minute over a 0.46x1.5m area (net width x 1.5m). Sample units in mineral substrates were obtained by dragging a 5cm-layer of sediment with the same net, over a 0.46x1.5m area. Prior to dragging, coarse substratum (pebbles) were brushed in front of the net, and then removed. The samples were preserved in the field in 4% formalin (final concentration). Invertebrates were sorted in the laboratory and preserved in 70% ethanol. They were mostly identified to Family (except for Annelida, Hydracarina, Nematoda and Planaria) and counted.

IV.2.3. Stream types, Least Impaired River Reaches (LIRRs) and Impaired River Reaches (IRRs)

The ratios between observed biological parameters and the expected values under reference conditions (Ecological Quality Ratios, see below) for these parameters are at the heart of WFD guidelines to evaluate river health (Hering et al., 2006). Hence, both typology and reference conditions need to be agreed upon before considering further developments. Chandesris and Wasson (2005) have established a typology of FG watersheds based on geomorphological, hydrological, and climate data. A biological typology of streams based on benthic macroinvertebrates (Dedieu et al., 2015) confirmed Chandesris' conclusion that small streams can be clustered into 2 major sub-regions (i.e. into 2 stream types), namely the "coastal alluvial plain" characterized by recent sediment and low elevations, and the inland "Guiana Shield" characterized by an eroded rocky substrate, a variability of elevations and large stream systems under a dense forest coverage.

Within each sub-region, we defined the status of each site (LIRRs vs. IRRs) based on National Survey Networks (DEAL Guyane, 2014), expert knowledge (e.g., presence of activities such as logging and gold mining was the most frequent criterion for IRRs), guidelines from the regional environmental agency, and recent biological and physical-chemical data collected by us (Dedieu et al., 2014, 2015). Mann-Whitney tests have been applied to compare physical-chemical characteristics between LIRRs and IRRs.

IV.2.4. Metric set

We considered 102 metrics. These metrics can be divided into 5 categories: (1) taxonomic richness-related metrics (e.g. the number of species in a particular taxa group or a combination of taxa), (2) abundance-based metrics (e.g. number of individuals), (3) diversity indices combining (1) and (2), e.g. Shannon's entropy; (4) functional metrics (e.g. feeding habits); and (5) tolerance-related metrics (e.g. Average Score Per Taxon, Armitage et al., 1983). For each site, each metric was calculated on the basis of (i) samples taken on organic substrates (A), (ii) samples taken on mineral substrates (B), and (iii) all samples (A+B) (Appendix V). Biological traits are poorly documented in stream invertebrates of FG compared to the European ones, so we selected five traits which are known at the family level for our fauna: functional feeding groups, locomotion, respiration, dispersal and habitat preferendum (Merritt and Cummins, 1996; Buss et al., 2004; Tamanova et al., 2006, 2008; Arrington and Winemiller, 2006; Ligeiro et al., 2010; Salman et al., 2013). Tolerance to pollution was based on the literature, e.g. Elmidae and Ecnomidae were considered as sensitive taxa following Couceiro et al., 2007 and Lorion and Kennedy, 2009. To quantify the sensitivity of taxa to water quality, the weighted average Chemical Pollution Index (CPI) of each taxa was calculated in order to determine the optimum value for a taxa (Ter Braak and Prentice, 1988). These values ranged from 0 (low sensitivity to water quality degradation) to 5 (high sensitivity, see Appendix V) and were used to calculate the Average Score Per Taxa (ASPT).

IV.2.5. Standardized Effect Size (SES) normalization, reference and worst values

In order to compare metric values obtained from different stream types, observed metric values were transformed into normalized deviations from values in reference conditions for a given stream type (Standardized Effect Size (SES); see Gotelli and McCabe, 2002). SES normalization allowed us to identify the direction of deviation from values in LIRRs and allowed a direct comparison of metrics, regardless of river typology.

SES values were calculated as follow:

$$\mathbf{SES = (Metric\_obs - mean_{ref})/sd_{ref}}$$

'Metric_obs' is the observed value of the metric, 'mean$_{ref}$' and 'sd$_{ref}$' are the average and standard deviation of the metric distribution under reference conditions for the same stream type.

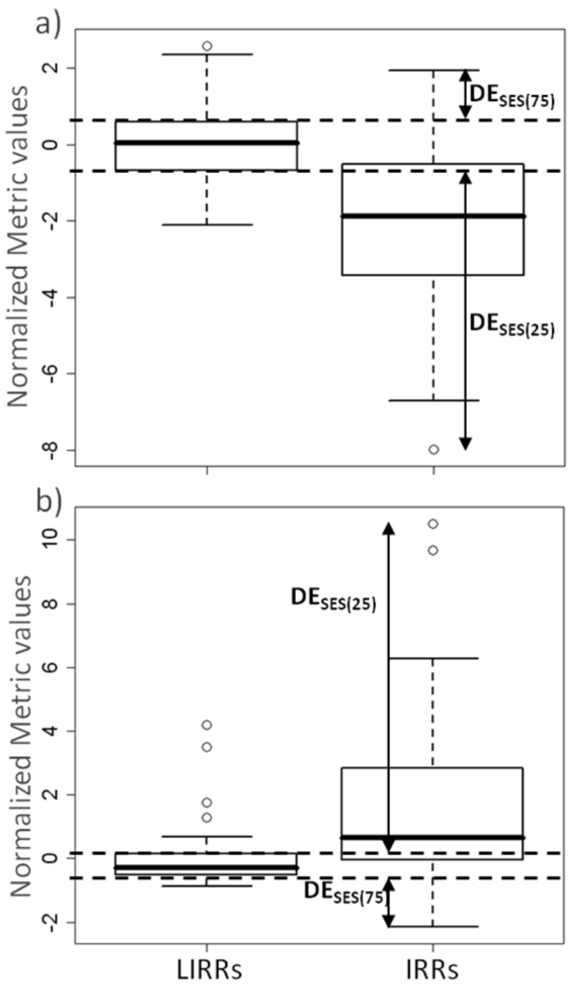

Figure. 32: Boxplots of Standardized Effect Size (SES) values of metrics in Least Impaired River Reaches (LIRRs. left box) and Impaired River Reaches (IRRs. right box). (a) Discrimination efficiencies (DEs) of a type 2 metric (Taxa Richness); (b) DEs of a type 3 metrics (GOLD Index). Boxes delineate the 25th and 75th quartiles; thick lines represent the medians; circles are extreme values; whiskers extend to maxima and minima.

Taking into account the discrimination efficiency of each variable (DE, Ofenböck et al., 2004), we determined the type of response of each metric in impaired conditions. A given metric could exhibit three response patterns: (i) not responding significantly to the impairment (type 1), i.e. the distribution of values from IRRs assemblages was not different from the distribution of values from LIRRs assemblages, and so neither $DE_{SES(25)}$ (proportion of IRR values smaller than the first quartile of the LIRR values distribution) nor $DE_{SES(75)}$ (proportion of IRR values higher than the third quartile of the LIRR values distribution) were higher than 0.25 (ii) decreasing with increasing impairment (type 2) (i.e. when $DE_{SES(25)}>0.25$ and $DE_{SES(75)}<DE_{SES(25)}$), (iii) increasing with increasing impairment (type 3) (i.e. when $DE_{SES(75)}>0.25$ and $DE_{SES(25)}<DE_{SES(75)}$) (see Figure 32). Last, we determined the reference values for each stream type and the worst value of each metric. The reference value corresponded to the highest (type 1 or 2) or the lowest (type 3) value this metric could take in the LIRRs from a given stream type. The worst metric value corresponded to the lowest (type 1 or 2) or highest (type 3) value a metric could take in the IRRs from the whole data set. The 5th and the 95th percentiles of the distribution of values for a given metric were used as reference or worst values. This was done in order to eliminate extreme metric values (Ofenböck et al., 2004).

IV.2.6. Metric normalization

In order to identify similar patterns in metric responses to anthropogenic pressures for all stream types, and thus to facilitate the selection of metrics used at a large spatial scale, the observed values of the metrics were transformed into Ecological Quality Ratios (EQRs) between observed and reference conditions for the same stream type at a time. EQRs were calculated following Hering et al. (2006), using formula [I] for metrics of types 1 and 2 and formula [II] for metrics of type 3.

EQR = (obs-lower)/(upper-lower) [I]

EQR = 1-(obs-lower)/(upper-lower) [II]

with "obs": the metric value observed for a given sampling point; "upper" and "lower" correspond to the "best" and "worst" value for this metric in the same stream type. In equation [II] "Upper "and" Lower "correspond to the"worst"and"best"value of the metric. EQRs were bounded between 0 and 1. If observed values were greater than the best value (if quality is higher than the reference data set), the value of the EQRs was limited to 1. Conversely if the values were smaller than the EQR worst value (if quality is lower than the worst values), the value of the EQR was fixed as 0.

IV.2.7. Metric selection

Our aim was to determine metrics which best discriminate non-impacted sites from the impacted ones. We selected metrics which were expected to show the best trade-off between (i) high discrimination efficiency (DE), (ii) low specificity and (iii) high stability under reference conditions (Mondy et al., 2012). The DE of a metric was calculated as the proportion of IRRs assemblages with lower EQR values than the first quartile of the LIRRs values distribution. The stability of a metric in reference conditions (i.e. LIRRs) was assessed using the coefficient of variation (CV) of EQR values distribution from LIRRs assemblages. Robust estimates of DE and CV were obtained through a bootstrap procedure (mean of 100 calculations, each calculation using 60 % of the sites randomly selected from the data set). We first selected the metrics which simultaneously exhibited a high DE and a high stability in LIRRs (average DE $\geq$ 0.5 and average CV in LIRRs $\leq$ 1/3).

Since we aimed at building a generalist index, metrics with a low specificity were preferred, i.e. we selected metrics which were significantly correlated (linear regressions, α<0.05) to a high number of environmental variables reflecting site degradation. Thus, metrics significantly correlated to at least 6 water quality variables out of 17 were selected (see variables in Table I). To avoid redundancy, candidate metrics providing the same biological or ecological information (e.g. 'taxonomic richness in group A' and 'taxonomic richness in groups A+B') were gathered into homogenous groups. Only the metric with the lowest specificity and the highest DE for a given group of metrics was selected for possible inclusion in the multimetric index. Last, in order to limit the number of metrics, previously selected metrics were put into a correlation matrix. Metrics with >75% correlation were grouped by category (see paragraph Metric set) and we selected the best metrics (non redundant in terms of bio-ecological information, and higher DE) for each group.

Finally, the IBMG was calculated using the following the equation:

$$\mathbf{IBMG} = \frac{\mathbf{\Sigma(DEm \times EQRm)}}{\mathbf{\Sigma DEm}}$$

With IBMG : French Guiana Macroinvertebrate Biotic Index (Indice Biotique Macroinvertébrés de Guyane in French), DE_m : the discrimination efficiency of the metric 'm' and EQR_m : the value of the metric 'm'.

IV.2.8. Ecological class boundaries and test of the IBMG

In accordance with the WFD, we proposed five quality classes (i.e. 'high', 'good', 'moderate', 'poor' and 'bad' ecological quality). The identification of the ecological quality class boundaries was based on the distribution of the IBMG scores of the development data set. Most of the reference sites should be rated as good or very good in biological condition, as per WFD guidelines. The median and the minimum value of the IBMG distribution in LIRRs were considered as the 'high-good' and the "good-moderate" boundaries, respectively (Figure 33). The "bad-poor", "poor-moderate" and "good-high" boundaries were set using quartile and medians of the LIRRs and IRRs distributions (see Figure 33).

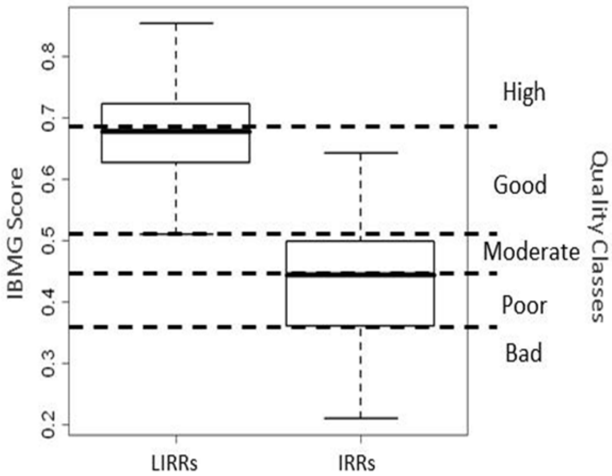

Figure 33: Ecological quality class boundaries (black dashed lines) for the IBMG. Boxplots represent the IBMG score distributions in Least Impaired River Reaches (LIRRs; left box) and Impaired River Reaches (IRRs; right box) ranging from the 25th to the 75th percentile of the distribution; thick lines represent the medians; circles are extreme values; whiskers extend to maxima and minima.

To evaluate the robustness of the IBMG discrimination efficiency, IBMG scores were calculated on the test data set (26 sites), and the DEs obtained with the development and the test data sets were compared. Then, the stability of IBMG values in LIRRs was tested using two Kolmogorov-Smirnov tests. These tests were used to evaluate significant differences in the distribution of IBMG values from LIRRs between the test sets and the development data set.
All statistical procedures were performed with R software (R Development Core Team, 2009).

IV.3. RESULTS

IV.3.1. Environmental variables

Regardless of stream types, some environmental variables differed significantly between LIRRs and IRRs. Compared to the reference sites, impaired sites had significantly higher values for turbidity, amount of total suspended matter, and % silt (Table IX). In the coastal alluvial plain, reference sites had lower total phosphorus concentration and % submerged vegetation than the impacted ones. Within the Guiana Shield area, impacted streams accumulated lower amounts of woody debris and leaf litter, and had lower percentage of submerged roots on the banks, compared to the reference sites. Water temperature was also slightly higher at these impacted sites, because of clearing of the riparian forest (Dedieu et al., 2014).

*Table IX: Physical-chemical characterics of the two stream types encountered in French Guiana. MN = Mean, SD = standard deviation. Note: NS indicates no significant difference; *p < 0.05 and ** p < 0.01 (Mann-Whitney tests).*

	Coastal alluvial plain					Guiana shield				
Variables	LIRRS		IRRS		*P*	LIRRS		IRRS		*P*
	MN	SD	MN	SD		MN	SD	MN	SD	
% Silt	8.04	9.04	48.24	21.43	**	8.61	6.82	34.00	20.63	**
% Sand	61.88	29.14	36.47	21.49	**	38.06	27.18	15.00	20.87	*
% Gravel	23.66	23.77	12.35	15.32	NS	19.12	15.45	19.17	14.78	NS
% Coarse Substratum	6.88	10.98	2.94	5.32	NS	34.17	27.29	32.67	26.58	NS
% Woody debris	16.34	14.34	14.71	11.52	NS	20.14	19.45	7.33	6.08	*
% Submerged vegetation	10.45	11.39	20.15	21.02	*	1.67	4.11	1.67	3.62	NS
% Leaf Litter	24.02	21.20	25.29	14.06	NS	17.89	13.89	13.83	17.85	*
% Submerged roots on the banks	20.26	16.51	9.26	7.33	NS	24.61	23.06	7.83	8.81	*
pH	5.29	0.54	5.33	0.31	*	6.19	0.79	6.38	0.38	NS
Water temperature (°C)	24.76	1.21	25.49	1.22	NS	24.36	1.63	25.64	1.91	*
Conductivity ($\mu S.cm^{-1}$)	22.25	5.05	26.47	15.24	NS	39.88	29.28	48.93	31.99	NS
Dissolved Oxygen ($mg.L^{-1}$)	6.02	0.93	5.71	0.85	NS	6.96	0.58	6.84	0.59	NS
Turbidity (NTU)	2.06	1.24	6.52	8.60	*	3.16	2.23	54.70	92.20	**
Nitrate ($\mu g.L^{-1}$)	0.30	0.15	0.24	0.12	NS	0.23	0.14	0.30	0.13	NS
Total phosphorus ($\mu g.L^{-1}$)	0.02	0.02	0.06	0.04	*	0.04	0.03	0.02	0.02	NS
Total suspended matter ($mg.L^{-1}$)	6.32	6.09	12.03	14.68	*	3.19	2.05	58.79	143.38	*
Dissolved organic carbon ($mg.L^{-1}$)	22.04	16.69	20.31	11.64	NS	13.23	9.87	12.27	6.23	NS

IV.3.2. Metric selection and index construction

Among the 102 metrics tested on the different sample groups (A, B, A+B), 56 differed significantly between LIRRs and IRRs in at least one sample group (Appendix V). Then, thirty-six metrics exhibited a mean DE greater than 0.5 and an average CV in LIRRS ≤ 1/3. (Table X). Seventeen of these 36 metrics significantly responded at least to 6 environmental variables (out of 17). From these 17 metrics, we eliminated redundant information by selecting the most discriminant metrics among the ones which provided similar bio-ecological information. After considering pairwise correlations of metrics, 12 metrics were eliminated. The Shannon index exhibited a DE slightly inferior to 0.5 (DE= 0.47, CV= 0.10); it was however added to the final multimetric index to fulfil recommendations of our National environmental agencies. Hence, the IBMG was finally composed of six metrics: Chao1 (B), Log.Elmidae (A), Number of Coleopteran families (A+B), %collector-gatherer (A+B), %Ephemeroptera and Trichoptera (A+B), and the Shannon index (A+B). The DE and CV in LIRRs of the IBMG were 0.79 and 0.15 respectively. The best value of each stream type and the worst values of the data set needed to calculate the EQR of each metric are given in Appendix V

Table X: Mean discrimination efficiency (DE), mean coefficient of variation (CV) and responses of 36 candidates metrics to the 17 environmental variables listed in Table I. DE= Discrimination Efficiency; CV= Coefficient of Variation; N = number of environmental variables significantly correlated to the metrics.

Candidate metrics	Description	DE	CV	N
Margalef_B	Margalef index calculated on group B samples	0.79	0.19	3
Chao1_AB	Chao estimator 1 calculated on A & B samples	0.76	0.11	10
TaxaS_A	Taxa richness calculated on goup A samples	0.76	0.12	5
TaxaS_AB	Taxa richness calculated on A & B samples	0.73	0.11	10
Chao1_A	Chao estimator 1 calculated on group A samples	0.72	0.12	5
Chao1_B	Chao estimator 1 calculated on group B samples	0.69	0.28	7
TaxaS_B	Taxa richness calculated on group B samples	0.69	0.26	5
Log.Elmidae_A	Logarithm of the abundance of Elmidae calculated on group A samples	0.69	0.32	7
Margalef_AB	Margalef index calculated on A & B samples	0.69	0.18	10
Margalef_A	Margalef index calculated on group A samples	0.65	0.17	2
Brillouin_A	Brillouin index calculated on group A samples	0.63	0.13	7
SwimQ_AB	% swimmers calculated on A & B samples	0.63	0.32	5
CoGaQ_A	% collector-gatherers calculated on group A samples	0.61	0.17	6
ETQ_AB	% Ephemeroptera and Trichoptera calculated on A & B samples	0.60	0.25	2
Simpson_B	Simpson index calculated calculated on group B samples	0.60	0.28	2
EPTLQ_A	% Ephemeroptera, Plecoptera, Trichoptera and Lepidoptera calculated on group A samples	0.60	0.27	7
EPTQ_A	% Ephemeroptera, Plecoptera, Trichoptera calculated on group A samples	0.59	0.30	3
EPTLMQ_AB	% Ephemeroptera, Plecoptera, Trichoptera, Lepidoptera and Megaloptera calculated on A & B samples	0.59	0.24	6
ColeoS_AB	Coleoptera families richness calculated on A & B samples	0.59	0.25	6
TrichoS_AB	Trichoptera families richness calculated on A & B samples	0.59	0.17	4
ColeoS_A	Coleoptera families richness calculated on group A samples	0.59	0.26	1
CoGaQ_AB	% collector-gatherers calculated on A & B samples	0.59	0.17	9
ETQ_A	% Ephemeroptera and Trichoptera calculated on group A samples	0.59	0.30	2
EPTQ_AB	% Ephemeroptera, Plecoptera and Trichoptera calculated on group B samples	0.58	0.24	1
EPTS_B	Ephemeroptera, Plecoptera and Trichoptera families richness calculated on group B samples	0.56	0.12	0
Falpha_A	Fisher's alpha index calculated on group A samples	0.56	0.30	2
Brillouin_AB	Brillouin index calculated on A & B samples	0.54	0.16	5
PMS_A	Plecoptera and Megaloptera families richness calculated on group A samples	0.54	0.21	3
ClingQ_AB	% clingers calculated on A &B samples	0.54	0.32	3
ShannonH_A	Shannon index calculated on group A samples	0.53	0.17	9
EPTLMS_B	Ephemeroptera, Plecoptera, Trichoptera, Lepidoptera and Megaloptera families richness calculated on group B samples	0.53	0.11	1
Simpson_A	Simpson index calculated calculated on group A samples	0.53	0.07	4
TrichoS_B	Trichoptera families richness calculated on group B samples	0.53	0.20	0
ETS_B	Ephemeroptera and Trichoptera families richness calculated on group B samples	0.52	0.12	1
TrichoS_A	Trichoptera families richness calculated on group A samples	0.51	0.19	1
ShannonH_AB	Shannon index calculated on A & B samples	0.47	0.10	5

IV.3.3. Ecological quality class boundaries and index test

The values of the IBMG scores of the development data set ranged from 0.21 to 0.85, and were used to set the quality class boundaries (Figure 33). The "good" lower boundary was set at 0.51 so that all reference sites were included in it (Barbour et al., 1999). The high-good and moderate-poor boundaries corresponding to the medians of the LIRRs and IRRs distributions and were set at 0.69 and 0.45 respectively. The poor-bad boundary was set at 0.36 corresponding to the 25th quartile of the distribution of IRRs scores. Values for the DE and CV in LIRRs calculated with the test data set were 0.83 and 0.11 respectively (Figure 34). The distributions of IBMG scores in LIRRs of the development and test data sets showed no significant difference (Kolmogorov-Smirnov test: $D = 0.1913$, $p = 0.8708$).

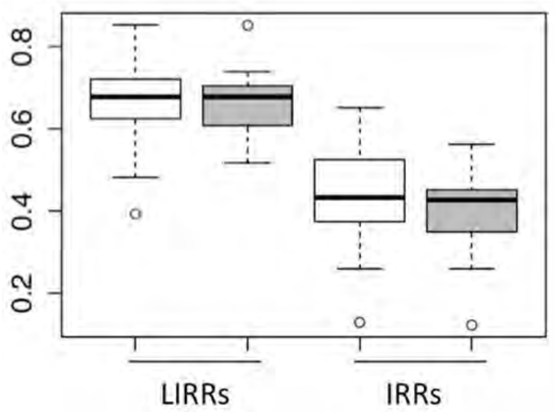

Figure 34: Distribution of the IBMG scores in Least Impaired Reaches (LIRRs) and Impaired River Reaches (IRRs) for the development data set (white boxes) and the test data sets (grey boxes). Rectangles delineate the 1st and 3rd quartiles; the thick lines represent the medians; circles are outliers: bars are maximal and minima.

IV.4. DISCUSSION

In this study, we propose a multimetric index for the biological assessment of East-Amazonian streams, developed under a EU framework. We assembled metrics that respond to several environmental variables associated to ecosystem impairment. Therefore, the IBMG can be considered as a generalist index in that it responds to the main types of pressures encountered in FG. Some studies showed that there is no significant difference in assemblage structure between dry and rainy seasons (Baptista et al., 2007; Couceiro et al., 2012). For practical reasons, the IBMG is based on samples taken during the dry season. Nevertheless, most remote sites cannot be reached (and therefore monitored) during the rainy season, and environmental managers will chiefly implement bioassessment campaigns during the dry season only. The IBMG can also be viewed as a preliminary tool in that it is based on a single campaign, and the test data set is limited to 26 sites. However, future surveys are expected to provide further data on the same network of sites during the next years, and this will allow to test and refine the index.

The six selected metrics account for effects of anthropogenic impairment on different environmental factors. Log.Elmidae and %collector-gatherers responded to changes in %woody debris, % submerged vegetation and % submerged roots, a series of organic habitat variables that indirectly account for the riparian habitat quality (Compin and Céréghino, 2007). Chao1, Shannon index, the % of Trichoptera-Ephemeroptera and the number of coleopteran families mostly correlated with mineral particle size (%sand, %gravel, %coarse substratum) and water chemistry (turbidity, total suspended matter, total phosphorus). These metrics therefore did well at accounting for changes in the instream physical-chemical habitat quality.

The fact that both A and B sample groups are included in the calculation of most metrics evidences that the structure of macroinvertebrate assemblages varied at the reach scale and that this variability is relevant for identifying the impaired status of streams in FG (Buss et al., 2004). Two metrics out of six are calculated on a group of samples only (A or B), thus reducing variability in the reference condition samples (LIRRs). Although, Chao1 estimator on sample group A+B had a higher DE and exhibited higher stability in reference conditions, it finally appeared to be highly correlated with the other candidate metrics. We thus selected Chao1 estimator on sample group B instead. Our index performed well in separating reference and disturbed sites since 79% of the sites that were a priori classified as impaired were assigned to categories below "good" quality by the IBMG. Testing the index with an independent data set also allowed us to demonstrate its stability in LIRRs and its robustness in terms of discrimination efficiency. Still, future works could improve the IBMG. Specifically, metrics able to detect diffuse pollution from the sparse human settlements or small cultivations are probably lacking, but these particular pressures are scarce in FG and could not be found with a sufficient replication in this study.

In other neotropical countries, multimetric indices were recently developed under National frameworks (summarized in Table XI). Interestingly, we note differences in selected metrics among countries. These differences might therefore be due to: (i) biogeographic differences in community structure, (ii) differences in anthropogenic pressure types and/or intensity, and/or (iii) differences in methods to design a multimetric index. Assuming that the Reference Condition Approach (RCA, Bailey et al., 1998) has become a worldwide standard to design biotic indices, one may expect that biogeographic differences in species composition and traits, and to some extent differences in anthropogenic pressures, primarily account for differences in metrics assemblages across countries.

Table XI: Comparison of multimetric indices recently developed in the neotropics under the reference condition approach. [1]Wastewater treatment plants; [2]Biological Monitoring Working Party; ID: Identification.

Reference	Region ; stream types	Study area (km²)	Main disturbance type(s)	Metrics selected	ID level
This study	French Guiana; 1st-2nd order streams	83 534	Gold mining; deforestation	Chao1 estimator Shannon index Coleoptera richness Log(Elmidae abundance) % collector-gatherers % ET	Family
Touron-Poncet *et al.* (2014)	Martinique and Guadeloupe islands; 1st-3rd order streams	1718	Urban, domestic and industrial runoff; WWTP[1] ; agriculture	Preference for silt Preference for boulders Abundance of Ephemeroptera ETC richness Trichoptera Richness Shannon index Taxa richness	Genus and species
Villamarin *et al.* (2013)	Ecuador and Peru; 1st -3rd order streams	1 568 736	Agriculture, livestock grazing; urban sewage	Richness of intolerant taxa EPT richness % clingers % climbers % tolerant taxa Taxa richness	Genus
Helson and Williams (2013)	Panama; 3rd- 4th order streams	1050	Agriculture, urban sewage	Margalef index Shannon index % EPT % Trichoptera Chironomidae:Diptera ratio % Scrapers % Shredders	Genus
Couceiro *et al.* (2012)	Brazil, central Amazon; 1st -2nd order streams	< 1000	Deforestation, urban sewage	Taxa richness (Family) EPT richness % EPT EPT:Chironomidae ratio Richness of sensitive taxa % collector-gatherers % Shredders	Genus
Moya *et al.* (2011)	Bolivia; 1st-4th order streams	1 098 581	Urban sewage, agriculture, mining, deforestation	Taxa richness (Family) EPT Richness % deposit feeders % microphyte feeders % coarse detritus feeders % flat-bodied taxa Total abundance % EPT	Family
Baptista *et al.* (2007)	Brazil, Southeastern Atlantic forest; 1st -3rd order streams	12 904	Urban sewage, agriculture	% Diptera % Coleoptera Taxa richness (Family) EPT Richness BMWP[2] index (adapted) % Shredders	Genus
Oliveira *et al.* (2011)	Brazil, Rio de Janeiro State; 1st-5th order streams	1265	Urban sewage	Taxa richness (Family) Trichoptera richness Shannon index % Plecoptera % EPT % Mollusca+Diptera % Shredders Hydropsychidae:Trichoptera ratio Chironomidae:Diptera ratio (%)	Genus
Ferreira *et al.* (2011)	Southeastern Brazil; 3rd- 4th order streams	29 173	Industry, mining, urban sewage, damming	Taxa richness (Family) % Oligochaeta % Chironomidae+Oligochaeta % EPT % collector-gatherers BMWP[2] index (adapted)	Family
Suriano *et al.* (2011)	Brazil, Sao Paulo State; 1st-2nd	248 800	Urban sewage, agriculture	Family richness EPT richness % EPT % Megaloptera+Hirudinae Shannon index (genus level) BMWP[2] index (adapted)	Genus

Taxonomic resolution is also an important issue in biological assessments (Waite et al., 2004) and most if not all rapid bioassessment methods rely on family or family-genus identifications of benthic macroinvertebrates (Heino et al., 2014). In recent neotropical studies, only Touron-Poncet et al. (2014) used species-level identifications of aquatic insects in Carribean islands – but in this insular context, most genera only had one species. On the methodological side however, multimetric indices in the neotropics were designed for study areas ranging from a few hundred (Couceiro et al., 2012) to about 1 million km^2 (Moya et al., 2011; Villamarin et al., 2013), and for stream types ranging from 1st-2nd order (this study) to 1st- 5th order (Oliveira et al., 2011) (see Table III). Differences in spatial and longitudinal scales have a significant influence on the natural variability of communities, thereby fostering the selection of certain metric types. For instance, while particular taxa are more likely to respond to disturbance at small geographic scales (e.g., Baptista et al., 2007; Touron-Poncet et al., 2014), a larger number of metrics that are independent of taxonomy (e.g. functional groups, biological traits) should theoretically be selected at larger scales (see Moya et al., 2011). Biogeography-related differences in final metrics assemblage can also be detected. For instance, Plecoptera richness or relative abundance commonly form metrics either per se or in combination with other sensitive taxa (Trichoptera, Ephemeroptera). This indicator group is not relevant in most of the neotropics where only one genus (Anacroneuria) is known (Fenoglio and Rościszewska, 2003), and in Carribean islands where it is entirely absent. These examples show that within a vast biogeographical area such as the neotropics, biological indicators developed for a given sub-area or country cannot be easily transposed to other geographic areas. Considering the influence of pressure types on assemblages of metrics, taxa that indicate water pollution (Chironomidae, Oligochaeta, Hirudinae) were more commonly selected in areas subjected to urban and agricultural pollution (e.g., Ferreira et al., 2011; Suriano et al., 2011), two types of disturbance that are infrequent in FG. Conversely, taxa with very specific requirements as regards mineral particle size do well at revealing degradation of the physical habitat (gold mining, deforestation in FG). This is particularly true of Trichoptera, especially in case-building taxa which require sand and piece of woods to build their larval case. Such selected examples support the hypothesis that freshwater Neotropical communities do not only depend on water quality but also of habitat components.

The IBMG fulfills the WFD requirements of taking into account the abundance and diversity of taxa. The inclusion of biological traits is highly desirable (Mondy et al., 2012), but unfortunately biological information is lacking in tropical areas where the autoecology of most species is poorly (or not) documented (Tomanova, 2007; Moya et al., 2011). Still, the IBMG includes a trait-related metric. Another requirement is that ecological evaluation should regard sensitivity of taxa to pollution (European Council, 2000). To date, tolerance values are lacking for the regional fauna and several studies have cautioned that tolerance values in the temperate regions do not apply to the neotropics (Moya et al., 2007). Initially, metrics indicating tolerance of taxa were calculated (see method section), but they were not selected by our statistical procedures. Nevertheless, Ephemeroptera, Trichoptera, and to a lesser extent Coleoptera are considered as sensitive to pollution by stream ecologists. Thus, the use of these taxa in our metrics, at least partially take into account "pollution sensitivity" of taxa within assemblages. Therefore we can reasonably consider that the index matches this last WFD criterion. Finally, comparing the IBMG with other indices reveals that, for several non-mutually exclusive reasons, multimetric indices developed in the neotropics may perform well in the context of the datasets used to generate them, but would certainly fail to be robust when used elsewhere. Ideally, there would thus be a need to intercalibrate these indices in an attempt to harmonize operational practices and reach a biogeographic region-wide biological assessment scheme.

Chapitre V. Evaluation de l’impact de l’orpaillage sur les cours d’eau de tête de bassin de Guyane à l’aide des traits biologiques des éphémères

Ce travail a fait l'objet d'une publication dans la revue scientifique Ecological Indicators

Résumé

Les petits cours d’eau de Guyane française, comme dans d'autres pays sud-américains sont fortement impactés par l’exploitation aurifère. La production annuelle d'or au niveau de la Guyane est 60 fois plus élevée qu'il y a 25 ans. Compte tenu de l’évolution économique actuelle et du cours de l’or constamment grandissant, il est urgent de développer des outils de bioévaluation robustes afin que les objectifs liés à la Directive Cadre sur l’eau puissent être établis pour la sauvegarde de la qualité des écosystèmes. Une approche basée sur la sensibilité des organismes à la pollution (saprobie) n’a pas pu être intégrée à l’indice que nous avons développé pour la Guyane. Les Éphéméroptères forment l'un des ordres les plus diversifiés dans la région néotropicale et des efforts pour améliorer la taxonomie de ce groupe ont été réalisés au cours des 20 dernières années en Amérique du Sud. Ces études permettent la potentielle utilisation de cet ordre, connu pour sa grande sensibilité aux perturbations humaines, pour le développement d'outils de bio-surveillance dans la région.

Les Éphéméroptères ont été échantillonnés sur 19 petits cours d’eaux dont 14 soumis à l’exploitation aurifère. Nous décrivons la composition en traits des communautés d’Éphéméroptères identifiés au niveau générique au travers de cinq traits et 21 modalités afin d’évaluer leurs réponses à la dégradation de l'environnement et de la chimie de l'eau causée par l’orpaillage. Nous avons observé un changement significatif de combinaison de traits entre les sites actuellement et anciennement soumis à l’exploitation aurifère et les sites de référence. Ces changements sont liés à des changements dans la richesse et la composition des communautés d’Éphéméroptères. Parmi les traits considérés, des changements dans le régime alimentaire, le mode de respiration et de locomotion ont été détectées dans les sites soumis à l’exploitation aurifère. Une analyse de correspondance floue a montré une ségrégation des sites actuellement orpaillés en fonction des traits des individus. Au niveau des sites anciennement orpaillés, aucune baisse significative des indices de diversité n’a été observée par rapport aux sites de référence, alors que la composition taxonomique et des traits étaient différents au niveau de ces sites. Ces résultats soutiennent donc le besoin de plus amples études sur la quantification des traits de cet ordre, pour une possible utilisation pour la bioévaluation de la qualité des cours d’eau néotropicaux.

Assessing the impact of gold mining in headwater streams of Eastern Amazonia using Ephemeroptera assemblages and biological traits

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Keywords: Biological traits; biomonitoring; fuzzy-coding; mayfly; neotropical rivers; macroinvertebrates

Summary

Whilst the biological traits composition of invertebrate assemblages has been successfully used to monitor temperate rivers, it has been seldom tested in tropical areas. We compared the trait composition of Ephemeroptera assemblages (five traits, 21 modalities) in three categories of headwater streams of FG: reference (undisturbed) sites, sites formerly impacted by gold-mining, and sites currently impacted by gold-mining. Differences in macroinvertebrate assemblage according to environmental characteristics and disturbance were evaluated using correspondence analysis and MANOVA. Among the considered traits, food acquisition, respiration and locomotion detected both past and current disturbance associated with gold-mining in headwaters. A fuzzy correspondence analysis showed a significant segregation of currently gold-mined, formerly gold-mined, and reference sites according to species traits. Shifts in trait composition were mostly related to changes in assemblage composition. Interestingly, no significant decline in diversity indices was observed in formerly gold-mined sites compared to the reference sites, 2 years after abandonment, while the taxonomic and trait composition of communities changed at these sites. These results support the case for further fundamental quantification of species traits, and for the inclusion of sensitive, trait-related metrics in upcoming multimetric indices for the assessment of river health.

V.1. INTRODUCTION

Throughout the world, environmental legislation aiming at surveying, managing and protecting freshwater ecosystems relies on biological indicators of ecosystem health (Stoddard et al., 2008; Dos Santos et al., 2011). For instance, regional-national surveys of stream systems provide large volumes of site-specific data on biological communities and the associated physical-chemical environments (e.g., Harris and Silveira, 1999; Paulsen et al., 2008; Mondy et al., 2012). The ecological health of rivers is then defined in terms of deviation from a *reference* state where human impacts are almost null (Bailey et al., 1998). Biological traits of freshwater organisms (e.g. body size, feeding habits, etc.) are potentially more useful than taxonomic structure (species x abundance data) to detect patterns of deviation from reference conditions where different hydroecoregions (areas that differ by geology, climate, vegetation, and species composition) are covered (Bonada et al. 2006), though ecologists traditionally make use of taxa lists (e.g., Bernadet et al., 2013). Whilst species occurrence may have a strong stochastic element and local-regional validity only, traits reflect environmental conditions and may be shared among many species (Southwood, 1988; Statzner et al., 2001). Traits may give greater insight into habitat change (Dolédec et al., 1999; Statzner et al., 2004) and their determination generally requires less taxonomic expertise (Dolédec et al., 2000), so that it can be utilized where limited information is available, and/or for animal groups where taxonomic knowledge is limited.

Most applications of biological traits to bioassessment were developed in the temperate zone, and were based on benthic macroinvertebrates (e.g. Usseglio-Polatera et al., 2000; Gayraud et al., 2003; Statzner et al., 2005; Dolédec et al., 2006). This is owing to the fact that species traits are poorly documented in tropical invertebrates compared, for instance, to the European or North-American ones (Touron-Poncet et al. 2014). To the best of our knowledge, only a few studies used a biological traits approach in tropical rivers (Tomanova, 2007. Tomanova et al;, 2008**)** to assess how macroinvertebrate community functions change along gradients of anthropogenic disturbance. Recent efforts on the taxonomy of the South American Ephemeroptera (mayflies) have provided valuable information on the diversity of this insect order in the neotropics (Heckman, 2002; Salles et al., 2004; Dominguez et al. 2006; Chacón et al., 2009), and in addition, Ephemeroptera are recognized as relevant biological indicators, because of their sensitivity to a wide array of disturbance types (Landa and Soldan 1991; Buffagni, 1997). Their taxonomic richness and/or abundance have notably proven relevant parameters for the design of multimetric indices in temperate (Gabriels et al. 2010) and neotropical (Couceiro et al., 2012; Touron-Poncet et al., 2014) rivers. Ephemeroptera are usually present in all stream types and benthic microhabitats within stream systems (Sowa, 1975), and show high morphological and ecological differentiation among genera (Dominguez et al., 2006). With such ecological characteristics, one may expect that variations in the biological trait combination of Ephemeroptera assemblages effectively account for ecosystem alteration.

The aim of this study was to assess the extent of shifts in the biological trait composition of Ephemeroptera assemblages along a gradient of disturbance associated with gold-mining in French Guiana (FG), East-Amazonia. Gold is the most significant mineral resource in the Guiana Shield (FG, Guyana and Surinam) (Hammond et al., 2007). After the prospection of large rivers, gold industries are now focusing on smaller inland streams (Cleary, 1990; Hammond et al., 2007). Small streams represent 80% of all running waters in FG and exhibit high ecological quality; some if not most of them have never been impacted by any human activity. Sediment discharges related to gold-mining activities are known to largely exceed those generated by other land-use changes, such as deforestation or road-building (Bruijnzeel, 1993; Krishnaswamy et al., 2006) and this type of disturbance certainly has harsh impact on the river biota in the Guiana shield where smalls streams naturally exhibit low levels of suspended materials (Hammond et al., 2007). In light of recent economic development, our ability to identify relevant reference conditions (e.g., community traits, ecological functions) and effectively rate ecosystem health will undoubtedly contribute to the success of future management actions. Assuming that environmental conditions strongly constraint Ephemeroptera assemblages (Hanquet et al., 2004), we hypothesized that i) streams with similar habitat conditions host Ephemeroptera assemblages with similar combinations of traits, and ii) anthropogenic disturbance generates broad shifts in ecological functions as species with certain traits are eliminated or replaced by species with other traits. In order to test these predictions, data on the Ephemeroptera assemblages were collected in 19 headwater streams of FG (abundance matrix for 35 genera), then five biological traits were described for the first time using a fuzzy-coding method (trait matrix). Matrix multiplication and a fuzzy-coding analysis were used to weight traits by taxa abundance, and to investigate the spatial distribution of trait combinations in relation to the extent of anthropogenic impacts generated by gold mining.

V.2. MATERIAL AND METHODS

V.2.1. Study area and sampling sites

This study was conducted in FG, East Amazonia, from October to December 2012. The climate is tropical moist with 3,000 – 3,400 mm of yearly precipitation mainly distributed over 280 days. There is a major drop in rainfall (dry season) between September and December and another shorter and more irregular dry period in March. The maximum monthly temperature averages 33.5° C (32.1-35.8°C), and the monthly minimum averages 20.3°C (19.7-21°C). The sampled streams had a water depth < 1 m and a stream width < 10 m, and were located in the upstream part of the river continuum. Larger streams and rivers were not considered in the study in order to focus on comparable ecosystems.

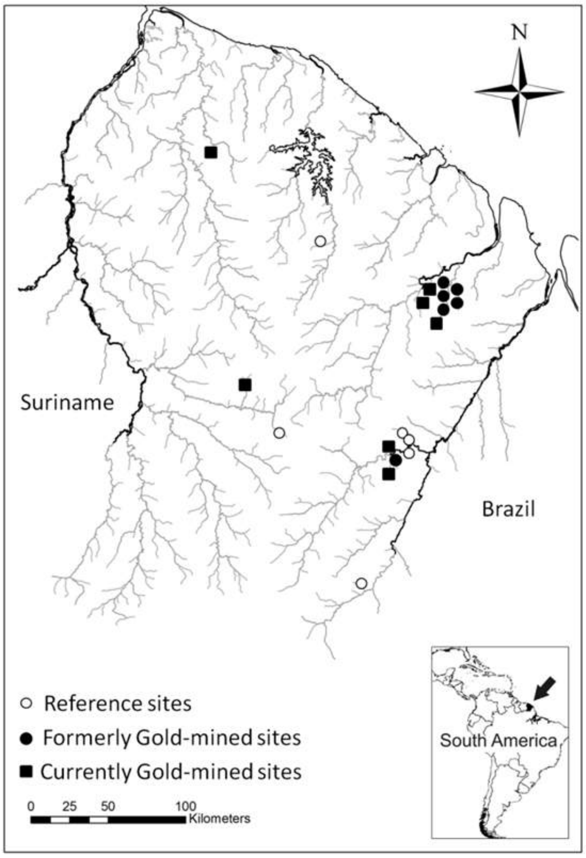


Figure 35: Distribution of 19 sampling sites in French Guiana

Nineteen sampling sites were sampled over 19 headwater streams belonging to FG's main river basins. Sites were mostly located on the northern part of FG, an area covered by dense rainforest and without road networks (Figure 35). The sites were sampled during the dry season (September-December) in 2012. Six sites were defined as not subjected to anthropogenic impacts (Reference sites). Impacted sites were currently (n=7) or formerly (n=6) subjected to gold-mining. Gold mining activity had stopped 2 years before sampling at the formerly gold-mined sites. The mining activities considered in this study refer to so-called "illegal" mining, i.e., small-scale traditional (or artisanal) mining which occurs in most South American countries (Hammond et al., 2007). These illegal activities involve small groups of workers (around a hundred people) who settle on small and remote forest streams (Hinton et al., 2003).

V.2.2. Physical-chemical variables

The length of a site was defined as 10 times its width, and transects were established each 5 metre along this length, for subsequent habitat measures. Stream flow (cm.s^{-1}) and the percentage composition of organic and mineral substrate types were determined on a 1 m^2 area every meter along each transect. Mean stream flow at a site was the mean of all point measurements. The substrate types included: %litter, %submerged roots on the banks, %macrophytes, %woody debris, %sand (particle size <2 mm), %gravel (2-25 mm), %coarse substratum (> 25 mm). Silt deposit being scarce naturally in FG streams, it was used to estimate the clogging of the river by gold mining.

We also measured chemical variables accounting for the impairment of stream ecosystem by human activities (Table I). Four variables were directly measured in the field using probes: dissolved oxygen (WTW 3205®), turbidity (EUTECH®131), pH (WTW 3110®) and conductivity (WTW 3110®132). Water temperature (°C) was the mean of values given by all above-mentioned probes. Following standard methods (AFNOR, 2005), total suspended matters (mg L^{-1}) were measured in laboratory based on water samples taken at each site and immediately frozen.

*Tableau XII: Physical-chemical characteristics of the three categories of sampling sites. Statistics are the results of Kruskal-Wallis test, comparisons made with values at the reference sites (*p < 0.05; ** p < 0.01; *** p < 0.001. ns: not significant).*

Variables	Reference	Formerly gold-mined sites		Current gold-mined sites	
	Mean±SD	Mean±SD	Statistics	Mean±SD	Statistics
Physical and water chemistry					
Temperature (°C)	24.96 ± 1.53	25.01±1.16	ns	25.52±2.13	ns
Conductivity (µs.cm-1)	32.2±24.69	35.1±19.01	ns	45.1±15.1	ns
pH	5.9 ± 2.59	6.01 ± 0.45	ns	6.04 ± 0.24	ns
Turbidity (NTU)	2.64±1.85	6.96±6.18	**	19.25±9.38	***
Total Suspended Matters (mg.L^{-1})	2.97±0.79	6.3±6.21	**	9.48±11.78	**
Dissolved Oxygen (mg.L^{-1})	6.97±0.35	5.21±1.45	**	6.53±0.85	ns
Mean stream flow (cm.s^{-1})	11.4±9.67	6.35±2.61	**	14.4±2.740	*
Substrates					
% Silt	13.33±6.83	45.5±15.08	***	44.16±14.970	***
% Sand	65±15.49	29.5±29.11	**	22.5±17.81	***
% Gravel	21.67±19.15	25±21.44	ns	34.16±19.6	*
% woody debris	21.51±10.65	18.31±30.48	ns	25.31±40.62	ns
% macrophytes	11.22±1.14	8.15±3.21	ns	1.59±3.11	***
% submerged roots on the banks	35.13±15.33	23.21±24.45	*	11.25±26.43	*
% litter	22.5±11.29	58.33±25.81	***	55.83±23.96	***

Significant differences in physical-chemical conditions were compared among reference, formerly gold-mined and currently gold-mined sites using the Kruskal–Wallis (K-W) test followed by *posthoc* pairwise comparisons (Wilcoxon’s test, hereafter W test) with a holm correction for multiple comparisons. Non-parametric tests were used because of small sample size and the heteroscedastic distribution of the assemblages.

V.2.3. Ephemeroptera sampling

Twelve sample units were taken at each site: 8 in organic substrates (roots, macrophytes, aquatic plants, litter, bryophytes) and 4 in mineral substrates (pebbles, gravels, sand, silt). Sample units in organic substrates consisted of intensive sweeping of a hand net (frame size=46 x 23 cm; mesh size = 500µm) through the substratum during 1 minute over a 0.46 m x1.5 m area (net width x 1.5m). Sample units in mineral substrates were obtained by dragging a 5 cm-layer of sediment with the same net, over a 0.46 m x1.5 m area. Prior to dragging, coarse particulates (pebbles) were brushed in front of the net, and then removed. The samples were preserved in the field in 4% (final concentration) formalin. Invertebrates were sorted in the laboratory and preserved in 70% ethanol. The individuals were sorted, identified and enumerated.

The Ephemeroptera were identified to the genus level using the key of Dominguez et al., (2006). Owing to the strong phylogenetic conservatism in the study traits of Ephemeroptera at the family and genus level (Merritt and Cummins, 1996), we considered the genus level as relevant for subsequent analyses of trait combinations.

V.2.4. Ephemeroptera assemblages and biological traits

First, correspondence analysis (CA) was used to ordinate the sites according to the hellinger-transformed abundance matrix (Legendre and Gallagher, 2001) for the 35 genera, thus summarizing the variability of Ephemeroptera assemblages, and providing insights for the discussion of the subsequent biological traits analysis. The significance of the axes was determined at $p<0.05$ by testing the eigenvalues of the inertia matrix. A permutational MANOVA (Anderson, 2001) was then used to evaluate differences in macroinvertebrate assemblage according to environmental characteristics and disturbance type. The significance of the test was given by F-tests based on sequential sums of squares from 1000 permutations of the data. As the order of non-orthogonal variables can have an effect on the outcomes of such method, the overall environmental descriptors of each microhabitat (habitat, substrates) and habitat (depth, width, velocity flow, pH) were first introduced in the analysis, and the mining-related variables (represented by turbidity and %silt) were the last considered. This approach was already implemented by Brosse et al., (2011) to take into account the pure effect of gold mining, in order to assess the effects of environmental characteristics and gold-mining intensity on fish communities of FG. Significant differences in genus richness and Shannon index were also evaluated among categories of disturbance, using K-W tests followed by *posthoc* pairwise comparisons (W test) with a holm correction for multiple comparisons.

The biological traits for each Ephemeroptera genus (Table XIII, Appendix VI) were obtained from the literature (Merrits and Cummins, 1996; Polegatto and Froehlich, 2003; Baptista et al., 2006; Dominguez et al., 2006; Tomanova et al., 2006, 2007), and the authors’ observations of live and preserved specimens (e.g. locomotion, food acquisition, mouthparts). The biological traits examined were likely to respond to two major environmental selective forces in stream ecosystems: habitat heterogeneity/stability (locomotion, maximum body size, body form, gill form), and food resource (feeding group) (Poff et al., 2006). The categories for each trait were either ordinal or nominal. Categories used for the different traits are listed in Table 2. Information on the biological traits was then structured using a fuzzy-coding technique (Chevenet et al., 1994): scores ranged from ‘0’, indicating ‘no affinity’ to ‘3’, indicating ‘high affinity’ for a given trait category. This procedure allowed us to build a trait matrix. We then used multivariate analyses to evaluate whether the distribution of traits in Ephemeroptera assemblages could discriminate reference, formerly and current gold mined sites. The trait matrix was first multiplied by taxa abundance in each site. The site-trait array was log-transformed into relative abundance of each trait category in each site and further processed by Fuzzy Correspondence Analysis (FCA) (Chevenet et al., 1994). We then examined if the first FCA axis (FCA1), which

Table XIII: Biological traits, categories (abbreviations as in Appendix VI) and functional trends (“+”: increase or “–“: decrease) along the disturbance gradient (sediment addition and altered hydrology) resulting from significant correlations between the proportion of trait categories and the first Fuzzy Correspondence Analysis axis (FCA1) axis scores.

Traits	Categories	FCA1	*p*-value
Gill Form	Leaf like gills (LG)		ns
	Filamentous gills (FG)	-	**
	Plate like gills (PG)		ns
	Numerous Tracheal filament (TG)	+	**
	Operculate gills (OG)	+	*
Body Form	Streamlined		ns
	Flattened	+	**
	Cylindrical		ns
Maximal Size	< 0.25 cm	-	*
	> 0.25 - 0.5 cm		ns
	> 0.5 - 0.7 cm		ns
	>0.7 cm	+	*
Feeding group	Brushers (Br)	-	**
	Scrapers (Scr)		ns
	Collector-Gatherers (CoGa)		ns
	Shredders (Shr)		ns
	Collector-Filterers (CoFi)	+	*
Mobility	Swimmers (Sw)	-	*
	Crawlers (Cr)		ns
	Epibenthic Burrowers (EpB)		ns
	Endobenthic Burrowers (EnB)	+	**

*P-values are for Pearson’s r tests: *p < 0.05; ** p < 0.01; ns: not significant.*

explained most of the variability in functional community composition, was related to the gold mining gradient. The relationships between FCA1 sites scores and the categories of impairments (reference, formerly gold-mined, currently gold-mined sites) were tested using a K-W test followed by *posthoc* pairwise comparisons (W test). Finally, Pearson's r coefficients were used to examine which trait categories were significantly correlated with FCA1 axis, in order to bring out the functional responses of taxa to anthropogenic perturbation.

All multivariate analyses and other statistical tests were implemented using the packages stats and ade4 (Chessel *et al*. 2004) in R 3.1.2 statistical software (R Development Core Team 2014).

V.3. RESULTS

V.3.1. Environmental variables

Physical-chemical variables (Table XII) differed significantly among the sites, according to the impairment type. Dissolved oxygen concentration and mean stream flow were significantly lower in formerly gold mined sites (K-W tests; *p*-value = 0.0023; *p*-value = 0.0093 respectively). Both currently and formerly gold-mined sites were characterized by higher turbidity and total suspended matters (K-W; Formerly : *p*-value = 0.0018; *p*-value = 0.0045; Currently: *p*-value = $2.4.E^{-5}$; *p*-value = 0.0021, respectively). The currently gold-mined sites had coarser substrates (gravels; K-W; *p*-value = 0.0089). The overall gold-mined sites (formerly and current) had more silt and less organic substrates (macrophytes and litter).

V.3.2. Responses of Ephemeroptera assemblages to gold-mining

The first and second CA axes described 19.01 % and 15.32 % of the total variability in Ephemeroptera assemblages respectively (Figure 36a). Such rather low explained variability is common when examining taxonomic assemblages (Fabrizi et al. 2010; Céréghino et al. 2012), while multivariate analyses of traits compositions provide much higher explanations of the total variability in the community structure (Dias et al., 2008; this study). A gradient of taxonomic structure matched a gradient of gold-mining activity along axis 2 (CA2) (Figure 36b), and the distribution of site coordinates along axis 2 differed significantly according to disturbance type (K-W; *p*-value: 0.000171 - see figure 2b for pairwise-comparisons.). Axis 1 of the CA thus rather displayed environmental variability within each category of site, suggesting a wide variation in assemblage among sites within a category (see e.g., reference sites, Figures 36a and 36b). Nevertheless, such within-category differences were due to uncommon (rare) genera, such as *Paramaka*, *Cryptonympha, Camelobaetidus, Terpides, Hydrosmilodon*. Some genera were, however, mainly found in gold-mined sites, namely *Hexagenia*, *Campusrus, Caenis, Apobaetids, Miroculis*, whereas other genera were associated with references sites (*Farrodes, Terpides, Rivudiva, Hagenulopsis).*

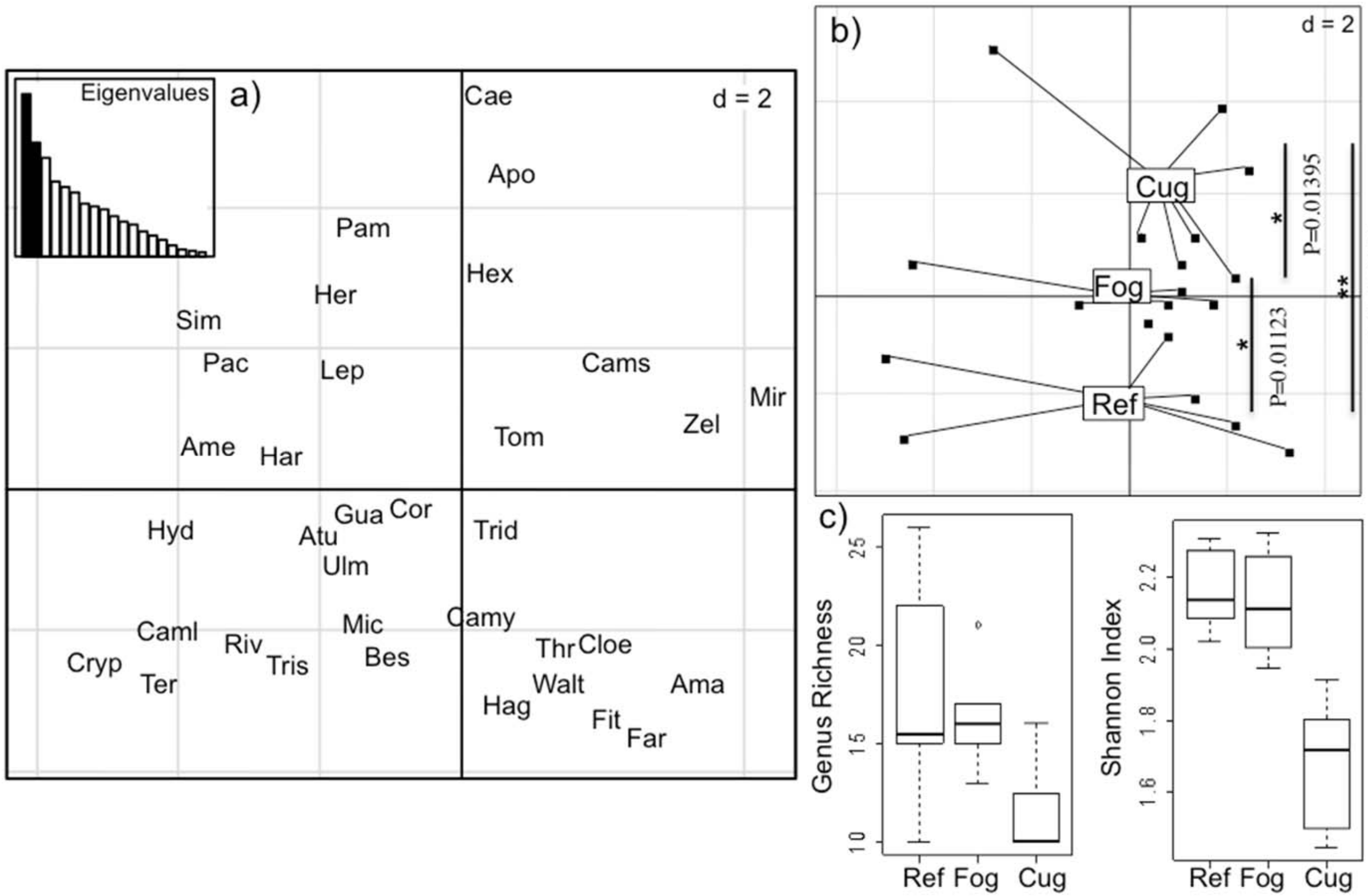


Figure 36. Correspondence Analysis (CA) of the taxonomic composition at the sampled sites. (a) Distribution of the genera in the first two factorial planes (See Appendix VI for labels of genera), (b) distribution of sites in the first factorial plans. The mean position of the three categories is at the weighted average of corresponding sites. Sites were linked to their corresponding category, (c) Genus richness and Shannon index of the different categories of disturbance (Ref: Reference sites ; Fog: Formerly gold-mined sites; Cug: Currently gold-mined sites).

Table XIV: Results of the MANOVA performed on Ephemeroptera abundance data

Source of variation	*MS*	*F*	r^2	*p*
Micro-habitat scale				
%sand	0.258	2.254	0.089	**0.020**
%litter	0.270	2.360	0.094	**0.012**
%roots	0.155	1.355	0.054	0.182
Habitat scale				
Width	0.152	1.330	0.053	0.233
Depth	0.099	0.865	0.034	0.507
pH	0.211	1.843	0.073	**0.048**
Stream Flow	0.145	1.262	0.050	0.267
Gold-minig				
Turbidity	0.243	2.123	0.084	**0.034**
%silt	0.207	1.806	0.072	0.077
Residual	0.115		0.397	

The significance of the tests was checked using F-tests based on sequential sums of squares from 1,000 permutations of the raw data (significant results are in bold).

A summary of the significance of the various effects of environmental parameters and gold-mining on the taxonomic structure of Ephemeroptera assemblages is given in Table XIV. 61% of the total variance of the Ephemeroptera assemblages was explained by our measured variables (permutational MANOVA, total R^2 = 0.603). Differences in Ephemeroptera composition between sites were significantly explained by %litter, %sand and pH (*p-values*: 0.020, 0.012 and 0.049 respectively). After accounting for the effects of stream habitat and micro-habitat descriptors, gold-mining still explained 8.2 % of the differences in assemblages between sites (*p-value*: 0.034). Moreover, significant differences across disturbance categories were apparent in terms of genus richness (K-W, *p*-value: 0.02902; Pairwise-Comparison Ref-Cug, *p*-value: 0.01339) and Shannon index (K-W, *p*-value: 0.001679, W, Ref-Cug, *p*-value: 0.003405; Fog-Cug, *p*-value: 0.003361) (Figure 36c).

V.3.3. Responses of Ephemeroptera traits to gold mining

The first and second FCA axes described 57.85% and 22.29% of the total variability in trait composition of assemblages, respectively. A gradient of biological traits clearly matched a gradient of gold-mining activity along axis 1 (FCA1) (Figure 37). Negative scores along FCA1 corresponded to reference and formerly gold-mined sites, whereas positive scores corresponded to the currently gold-mined ones. We found a significant difference of FCA1 coordinates between these categories of impairment (K-W; *p*-value: 0.00149 - see Figure 37b for pairwise-comparisons).

Some correlations between trait categories and FCA1 axis scores were statistically significant (Table XIII). Within assemblages, the increasing proportions of large individuals (up to 0.7 cm), flattened bodies, endobenthic burrowers, collector-filterers and individuals with operculate gills or with tracheal filaments were correlated with the gold-mining gradient (Figure 37a).

Conversely, small individuals (< 0.25 cm), brushers, swimmers and individuals with filamentous gills were preferentially associated to reference sites. We also noticed that formerly gold-mined sites were located on the middle area of the scatterplot, and that mid-sized organisms (0.5-0.7 cm), collector-gatherers, individuals with plate and leaf like-gills, crawlers, and epibenthic burrower seemed to be favored in these sites (Figure 37b – Table XIII).

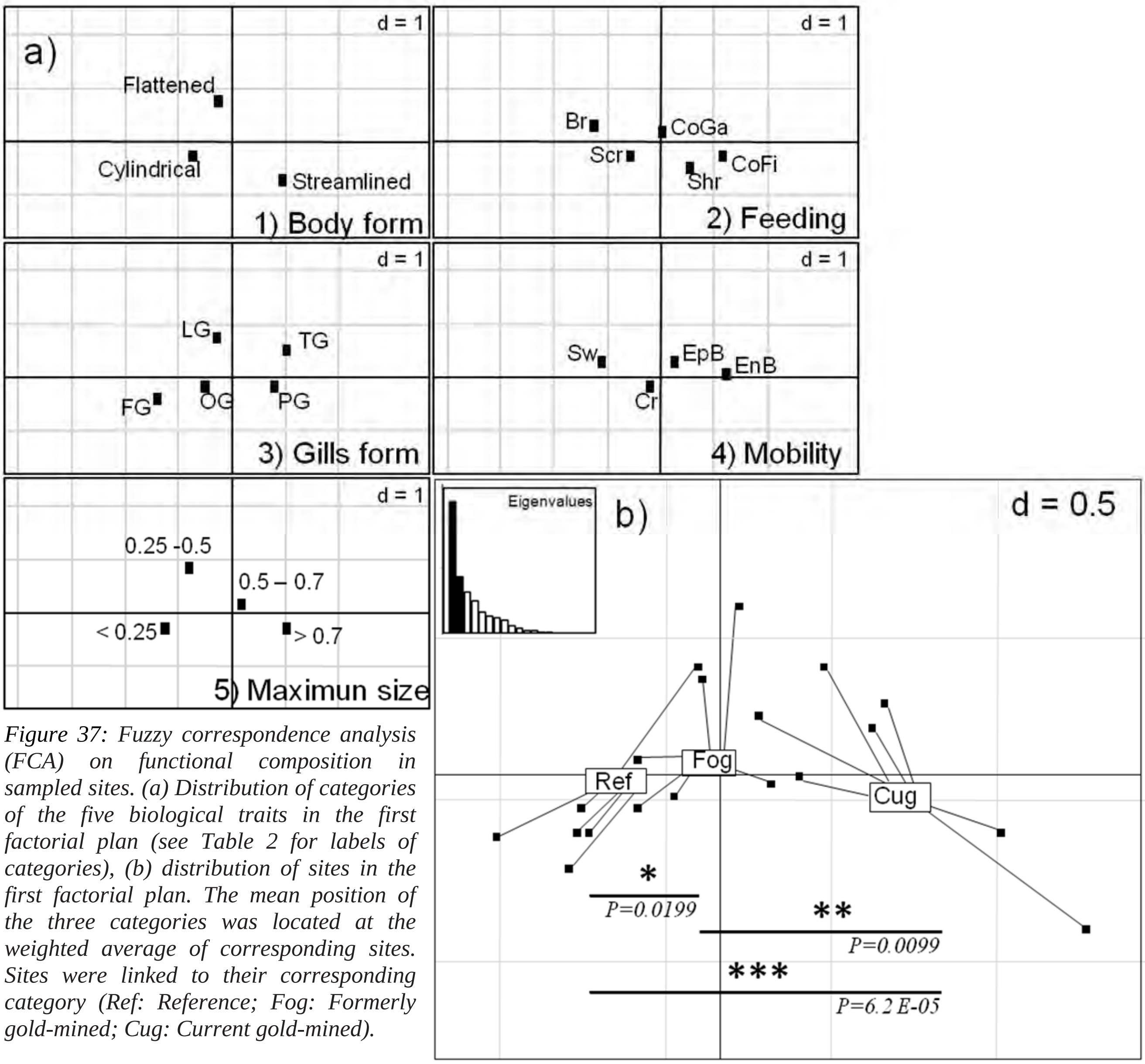


Figure 37: Fuzzy correspondence analysis (FCA) on functional composition in sampled sites. (a) Distribution of categories of the five biological traits in the first factorial plan (see Table 2 for labels of categories), (b) distribution of sites in the first factorial plan. The mean position of the three categories was located at the weighted average of corresponding sites. Sites were linked to their corresponding category (Ref: Reference; Fog: Formerly gold-mined; Cug: Current gold-mined).

V.4. DISCUSSION

Biological traits of macroinvertebrates were successfully used in previous studies to reveal impacts of various types of disturbance, especially in the temperate zone where biological attributes of the benthic fauna are well documented (e.g. Dolédec et al., 1999; Usseglio-Polatera et al., 2000; Gayraud et al., 2003; Statzner et al., 2004; Dolédec et al., 2006). We know that biological traits (life history patterns, body size, etc.), species richness and numerical dominance do not compare among biogeographic regions, thus precluding the transposition of associations of biological traits and environmental conditions from temperate to tropical areas. Nevertheless, once biological traits are quantified for a given ecoregion (e.g., this study), we can take advantage of the strong phylogenetic conservatism of traits at the genus-family level to broaden their application beyond the local context used to generate them. In addition to biogeographic differences in traits, it is worth noting that the dominant economic activities that generate strong impacts upon river ecosystems do not fully compare between tropical (e.g., gold mining, timber for wood product) and temperate localities (e.g., intensive agriculture, industry, urbanization). Gold is the most significant mineral resource in the Guiana Shield (FG, Guyana and Surinam), and as such, has driven mining activity for centuries (Hammond et al., 2007). Ephemeroptera traits based on food acquisition, respiration and locomotion did well at detecting both past and current disturbance associated with gold-mining in headwaters. There was a clear shift of trait combinations from currently gold-mined sites to the formerly gold-mined ones, and then to reference sites. In light of our results, we can state that shifts in trait composition were related to changes in Ephemeroptera assemblage composition and richness.

Sediment addition is probably the structuring factor for assemblages in gold-mined sites. Several studies previously demonstrated that discharges from mining activities decrease species diversity and alter species composition (e.g. Beltman et al., 1999 ; Malmqvist and Hoffsten, 1999 ; Soucek et al., 2000; Tarras-Wahlberg et al., 2001 ; Yule et al., 2010). Vasconcelos and Melo (2008) documented the short-term impact of sediment release on tropical macroinvertebrates diversity. We found that higher proportions of endobenthic burrowers and collector-filterers were associated to gold-mined sites. In our study, this combination of traits corresponded to the so-called burrowing mayflies from the genera *Campsurus* (Polymitarcyidae) and *Hexagenia* (Ephemeridae), which are well adapted to deposited substrates (sand, silt). These Ephemeroptera inhabit U-shaped tunnels burrowed into clay and feed by resuspending the organic particles (Merritt and Cummins, 1996; Dominguez et al., 2006). We also noticed that individuals with leaf-like or filamentous gills were preferentially associated to undisturbed sites. Filamentous gills are considered as the most fragile organs in the Ephemeroptera order and may be linked to the absence of particles in the water column. Conversely, operculate gills (e.g. *Caenis*) act as protective covers thus confering resistance to sediment increase. Gold-mining activities also lead to the loss of physical habitats such as macrophytes and mineral substrates. Habitats such as aquatic macrophytes provide direct (refuge habitat) or indirect (support for the development of algae and biofilm that constitute food) resource supplies (Allan, 2007). In reference sites, we mainly found herbivorous taxa (brushers and scrapers) feeding over the surface of coarse mineral particles. This result corroborates former studies in neotropical areas which showed that sediment release due to gold-mining clog the river bottom (Mol et Ouboter, 2004). Higher sediment loads into the watercourse lead to the decline of primary producers, and subsequently, the herbivores (e.g., the Baetidae in our study) that depend on them (Parkhill and Gulliver, 2002; Suren et al., 2005; Tudesque et al., 2012).

The above-discussed results suggest a strong relationship between habitat conditions and Ephemeroptera traits. However, some differences in Ephemeroptera diversity also accounted for variations in local environmental conditions. Regardless of human-induced perturbation, variations in % sand, % litter and pH were the most structuring factors. Where allochthonous inputs in the form of coarse organic matter are important (e.g. litter), it is expected that shredders should be the numerically dominant non-predators. At sites where autochthonous energy is more important, scrapers should have greater numerical importance. Most of our sites were located in forested areas, with a more or less dense canopy cover by overhanging tress. They thus exhibited variable accumulations of leaf litter over the bottom. Many of the dominant Ephemeroptera were associated with leaf debris, notably *Campylocia* (filterer-colllector), *Farrodes* (scraper) and *Waltzohyphius* (collector), thus explaining the importance of the variable % litter. Sandy substrates are typical microhabitats of the burrowing mayflies (see above). Ephemeroptera diversity is known to be adversely affected by low pH values (Bell, 1971; Sutcliffe and Carrick, 1973; Rowe et al., 1988; Courtney and Clements, 2000; O'Halloran et al., 2008). The genera *Camelobaetidius*, *Hermanella* and *Thraulodes* were only found in remote reference sites with higher pH (> 5.8). These results suggest that neotropical Ephemeroptera assemblages were partly shaped by deterministic processes (Lepori and Malmqvist, 2009). Finally, it is likely that the stochastic occurence of rare taxa like *Paramaka, Terpides, Hydrosmilodon* contributes to between-site variability in assemblage diversity in reference and impacted sites. It is worth noting that no significant decline in genus richness and Shannon entropy was observed in formerly gold-mined sites 2 years after abandonment, compared to the reference sites, but the taxonomic and trait composition of communities changed at these sites. Both traits and taxa compositions thus have the potential to reveal the persisting effect of the harsh physical modifications that once occurred with mining (e.g. deforestation, channelization, creation of artificial pools) on current benthic assemblages. Similarly, Brosse et al., (2011) found an incomplete resilience of fish assemblages after the abandonment of sites in FG, in areas where mining activities had stopped for one year.

However, we note that biological metrics such as richness or Shannon index, commonly used as basic indicators of ecosystem impairment, or even to design multimetric indices in the neotropics (Baptista et al., 2007; Helson et Williams, 2013; Villamarin et al., 2013; Touron Poncet et al., 2014) were inefficient at detecting the “ghost of past disturbance”. Assuming that current multimetric indices developed for neotropical streams do well at quantifying shifts in taxonomic structure, the inclusion of trait-related metrics could lead to relevant improvements of these biological assessment tools. Moreover, recolonization by Ephemeroptera can occur through the oviposition of aerial adults, upstream movement, downstream drift or movement upward from the hyporeic zone (Williams and Hynes, 1976). The contribution from these various sources of colonists will vary with species and situations (Townsend and Hildrew, 1976). Further works on the quantification of new traits, notably those related to life history patterns (e.g., number of generations per year), reproduction (e.g., number of eggs per female adult) and dispersal ability (drifting behavior, flight abilities) should therefore help to evaluate more precisely the effect of human disturbances on the ecological integrity of neotropical streams (Brittain, 1991; Dolédec et al., 1999, 2006).

V.5. CONCLUSION

Although the biological traits approach to biomonitoring has many benefits, its use in neotropical streams is still in its infancy. Beyond the need to document species traits through studies of individual and population biology, there will be a need to determine which traits best respond to which type of human impacts. For instance, Ephemeroptera assemblages may not be sensitive to a full range of potential disturbances encountered in eastern Amazonia, although they do well at detecting gold-mining impacts in this area. Traits of sensitive groups such as the Ephemeroptera could also be considered in multimetric indices that comply with the EU Water Framework Directive guidelines, while such biomonitoring tools are progressively set up and implemented in overseas region of Europe (Touron-Poncet et al., 2014; Dedieu et al. submitted). Lastly, our published trait profiles of Ephemeroptera will hopefully contribute to future trait approaches intended to compare ecological functions in pristine and impacted streams in nearby neotropical areas (Appendix VI).

Chapitre VI: L'indice biologique Macroinvertébrés de Guyane (IBMG)

Ce chapitre pourra faire office de guide méthodologique pour la mise en application de l'indice biologique

VI.1. L’IBMG ET LA DIRECTIVE CADRE SUR L’EAU

VI.1.1. Les exigences de la DCE

Nous avons constaté d’une manière générale un manque de connaissance sur l’écologie des macroinvertébrés benthiques de la Guyane Française, qu’il s’agisse de leur degré de sensibilité aux pollutions, de leurs préférences d’habitat, de leur distribution longitudinale suivant des gradients environnementaux naturels ou encore des traits biologiques des espèces. Compte tenu de ces observations et des objectifs fixés par la DCE, la démarche pour développer un indice basé sur le groupe des macroinvertébrés, adapté à la Guyane Française et DCE-compatible, a été la suivante :

1) Acquisition de données faunistiques et physico-chimiques chaque stations et pour chacune des deux campagnes de terrain ;
2) Réalisation d’une typologie des petits cours d’eau de Guyane Française sur la base de la faune. Cette étape permet de mettre en évidence des groupes de stations homogènes du point de vue de leurs communautés. Elle permet aussi de mettre en évidence les paramètres environnementaux naturels ou d’origines anthropiques structurant les communautés ;
3) Calcul des métriques descriptives des communautés, et normalisation en « écart à la situation de référence » (EQR) ;
4) Sélection des métriques pertinentes, et agrégation des métriques sélectionnées en un indice multimétrique final ;
5) Test de l’indice développé sur un jeu de données différent. Les limites des classes de qualité ont été établies, et l’état écologique de chaque site évalué.

Ces étapes ont abouti à la proposition de L’indice Biologique Macroinvertébrés de Guyane (IBMG).

VI.1.2. Typologie des petites masses d’eau

La variabilité environnementale a été principalement abordée sous la forme adoptée dans la DCE : les hydro-écorégions (Wasson et al., 2002). La définition de ces « **types de cours d’eau** » est basée sur des descripteurs de l’environnement majeurs tels que la géologie, l’altitude, la taille du cours d’eau (Chapitre I). Il est aussi reconnu que la définition d’une typologie de rivière permet une meilleure comparaison entre différentes communautés biologiques (Verdonschot et

Nijboer, 2004). L'objectif était donc de mettre en évidence des descripteurs simples ayant un sens écologique dans le contexte de la Guyane. Pour cela, les hydro-écorégions de Guyane (Chandesris et al., 2005 – Figure 38) ont été validées avec une approche *a posteriori* (analyse de groupement) afin de définir des cours d'eau « type » (Chapitre III).

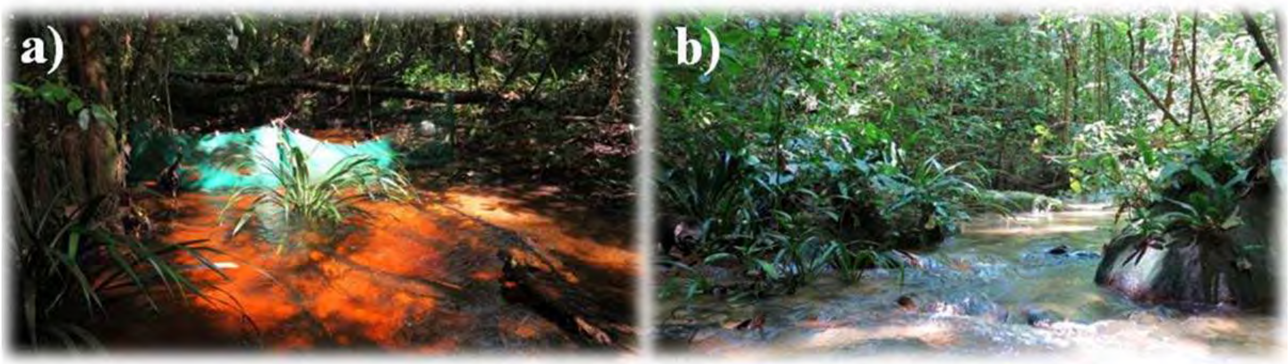


Figure 38 : Les deux types de PME : a) les PME de plaine alluviale et b) les PME du bouclier Guyanais.

Tout d'abord, une classification de 65 stations sur la base des invertébrés a permis de confirmer que **les HER résultaient en des différences dans les assemblages benthiques.** Ensuite, l'analyse des gradients de variables biologique (distribution des invertébrés) et abiotiques correspondantes nous a permis de caractériser les deux hydro-écorégions à partir de paramètres environnementaux simples. En combinant l'élévation, le pH et des variables d'habitats (granulométrie, macrophytes), une typologie des petits cours d'eau de Guyane a ainsi pu être proposée (Tableau XV). Ce tableau servira par la suite pour rattacher les sites échantillonnés à leurs hydro-écorégions respectives (Figure 11 et 38).

Tableau XV: Description des deux hydro-écorégions de Guyane Française

Hydroécorégions	Elevation	pH	Granulométrie	Macrophytes	N
Plaine alluviale	0 - 200 m	< 5.8	Sable majoritaire	Présent	51
Bouclier Guyanais	> 200 m	> 5.8	Substrat grossier	Rare	42

VI.2. L'IBMG

Les objectifs initiaux sont remplis puisque l'IBMG répond aux exigences de la DCE en tenant compte i/ des conditions naturelles pour exprimer le résultat de l'évaluation d'un site en terme d'écart à la référence pour le type de cours d'eau considéré, ii/ de la diversité et iii/ de l’abondance des taxons (Chapitre III). De plus, l’indice actuel a reclassé correctement 79% des stations du jeu test au sein de leurs catégories respectives.

VI.2.1. Sensibilité de l’IBMG

La majeure partie des sites perturbés échantillonnés au cours de cette étude correspondait à des cours d’eaux soumis à l’exploitation aurifère et forestière. Nous pouvons émettre l’hypothèse que l’indice développé soit actuellement plus sensible aux perturbations **physiques** (Sédimentation, MES, perturbation hydrobiologie) que chimiques (ex : pollution urbaine). L’indice nous semble toutefois pertinent pour les PME qui sont généralement situées en tête de bassin et donc plus soumises à ce type d’activité humaine. Cependant, avec la croissance démographique actuelle, il est possible que la population guyanaise s’étende plus profondément dans les terres. A l’avenir, l’impact de l’urbanisation (agriculture sur abatis, rejet domestique) devra être évalué plus particulièrement, d’autant que leur impact négatif sur les organismes aquatiques a largement été documentés en milieu tempéré (Cuffney, 2010; Beketov et al., 2013) et en zone tropicale (Castillo et al., 2006; Kleine et al., 2012 ; Couceiro et al., 2007). Même si l’IBMG s’adresse dans un premier temps aux têtes de bassin, les méthodes et routines statistiques développées dans ce travail permettront néanmoins une intégration rapide de données nouvelles en vue d’élargir la portée de l’outil.

VI.2.2. Les métriques biologiques

L’IBMG est composé de six métriques (Tableau XVI) : deux métriques basées sur la **richesse taxonomique** (Estimateur de Chao1, le nombre de familles de coléoptères), deux métriques d’**abondance** (Log.Elmidae, % d’Ephemeroptères et de Trichoptères), une métrique **fonctionnelle** (% collecteurs) et un **indice de diversité** (l’indice de Shannon).

Tableau XVI: Les six métriques composant l’indice biologique macroinvertébré de Guyane.

Code de la métrique	Métrique	Type de métrique	DE	Phase	Type de pression
Chao1_B	Estimateur de Chao1	Diversité	0.69	Minérale (4)	Habitat, eau
Log.Elmidae_A	Logarithme de l'abondance des Elmidae	Abondance	0.69	Organique (8)	Ripiscilve
ETQ_AB	Pourcentage de Trichoptères et d'Ephéméroptères	Abondance/Tolerance	0.60	Total (12)	Habitat, eau
CoGaQ_AB	Pourcentage d'individus collecteurs	Trophie	0.59	Total (12)	Ripiscilve
ColeoS_AB	Nombre de familles de Coléoptères	Diversité	0.59	Total (12)	Habitat
Shannon_AB	Indice de Shannon	Diversité	0.47	Total (12)	Habitat, eau

avec DE : Efficacité de discrimination ; Phase : Phase d’échantillonnage sur laquelle est calculée la métrique ; Type de pression : le type pression à laquelle la métrique répond.

VI.2.2.1. L’estimateur de Chao1

L’estimateur de Chao1 est un outil statistique robuste pour évaluer la richesse minimale d’un échantillon (Shen and Chao, 2003; Chao et al., 2009). Dans le cadre de l’estimation d’un nombre d'espèces présentes de type « asymptotique » (tendance vers un maximal théorique ou s'en rapprochant sans jamais l'atteindre), des échantillonnages supplémentaires sont souvent nécessaires pour identifier les espèces non détectés. Le principe de cet estimateur est que:

- Si une communauté en cours d'échantillonnage présente des espèces rares (singletons), alors elle possède encore des espèces à découvrir,

- Lorsque toutes les espèces auront été récupérés au moins deux fois (doubletons) dans l’échantillon, alors toutes les espèces présentes auront été détectées.

Cette métrique est particulièrement intéressante pour l’évaluation de la diversité de la phase minérale ou le nombre de prélèvement (n=4) est plus faible que pour la phase organique. L’estimateur de Chao1 se calcule de la manière suivante:

$$S_1 = S_{obs} + \frac{F_1^2}{2F_2}$$

avec Sobs : le nombre de taxon dans l'échantillon,
F1 : le nombre de « singletons » (le nombre d'espèces avec une seule occurrence dans l'échantillon)
F2 : le nombre de « doubletons » (le nombre d'espèces avec deux occurrences dans l'échantillon).

VI.2.2.2. Les Elmidae, le nombre de famille de Coléoptères et le pourcentage de Trichoptères et d’Ephéméroptères.

Les Ephéméroptères et les Trichoptères font partie des bioindicateurs les plus fiables et sont couramment utilisées pour leur sensibilité à la dégradation de l'environnement. Ces deux ordres composent de nombreux indices néotropicaux sous la forme de métriques individuelles (exemples : *Pourcentage de Trichoptères* (Helson et Williams, 2013), *Richesse en Trichoptères* (Oliveira et al., 2011 ; Touron-Poncet et al., 2014), *Abondance en Ephéméroptères* (Touron-Poncet et al., 2014)) et sont généralement identifiées à des niveaux taxonomiques plus fins (genre, espèce). Les avancées taxonomiques dans les pays limitrophes (ex. : Suriname : Flint, 1974 ; Brésil : Paprocki et al., 2002 ; Dominguez et al., 2006) permettraient probablement de déterminer un nombre de genre et d’espèce dont on pense qu’ils peuvent être communs (notamment au sein du bouclier Guyanais). A titre d’exemple, la comparaison avec un inventaire des Trichoptères au Surinam (124 espèces connues contre seulement en 4 Guyane Française (Flint, 1974)) démontre la carence aiguë de nos connaissances systématiques. L’ordre des coléoptères, représenté par deux métriques dans notre l’indice (richesse en famille et l’abondance d’Elmidae) semble aussi être un paramètre pertinent pour la bioévaluation en Guyane. Les coléoptères sont des organismes largement utilisés en bioindication (McGeoch, 2007*)* et sont de bon indicateur des caractéristiques de l'habitat écologique (Eyre et Foster, 1989; Ribera et Foster et al., 1992 ; Sanchez-Fernandez et al., 2004 ; Sharma et al., 2013). Les individus de la famille des Elmidae ont été rencontrés fréquemment dans les PME et nous avons observé que leur abondance diminuait dans les sites impactés par l’homme. Actuellement, 11 familles et 35 genres de coléoptères sont identifiés en Guyane (Museum National d’Histoire Naturelle - Jean-Philippe Champenoi, communication personnelle) mais nous suspectons que beaucoup plus sont présents.

VI.2.2.3. Le pourcentage d’individus collecteurs

Une métrique fonctionnelle « % collector-gatherer » (%collecteurs) à été intégrée à l’IBMG. Cette métrique s’avère particulièrement intéressante pour évaluer la qualité de la zone riveraine. En effet, la ripisylve fournit un apport de ressource important (ex : litière) pour les macroinvertébrés de type « collecteur » (Cummins et al., 1989; Sweeney, 1993). A l’inverse, l’abondance des macroinvertébrés de type « racleur» ou « herbivore» aurait tendance à augmenter dans les cours d'eau où le couvert forestier a été réduit suite au défrichement des forêts et cela principalement due à l'augmentation de la production primaire (Hawkins et al., 1982; Bojsen et Jacobsen, 2003). Les individus de type « racleur» et « herbivore » ayant été rencontrés en densité plus faible (à l’exception des Baetidae et des mollusques), la métrique liée au groupe « collecteur » a été retenue car elle discriminait mieux les sites perturbés.

VI.2.2.4. L’indice de Shannon

L’indice de Shannon est un indicateur largement utilisé pour l’estimation de la diversité biologique (Buckland et al., 2005). L’indice de Shannon correspond aux indices de type « I » qui donnent plus d’importance aux variations des espèces rares plutôt qu’au plus abondante (Peet, 1974). Cet indice permet ainsi d’exprimer la diversité en prenant en compte le nombre d’espèces ainsi que l’abondance des individus au sein de chacune de ces espèces. Ainsi, une communauté dominée par une seule espèce aura un coefficient moindre qu’une communauté dont toutes les espèces sont codominantes. La valeur de l’indice varie de 0 (une seule espèce ou bien une espèce dominante) à log S (lorsque toutes les espèces ont même abondance). Cet indice est d’une utilité relative puisque la composition d’une communauté peut changer sans faire varier la diversité, cependant, sa combinaison avec d’autres mesures biologiques est pertinente en écologie (Magurran, 1988). L’indice de Shannon se calcule de la façon suivante :

$$H' = -\sum_{i=1}^{S} p_i \log_2 p_i$$

avec: i : un taxon du milieu d’étude.

p_i : Proportion d’un taxon i par rapport au nombre total de taxon (S) dans le milieu d’étude qui se calcule de la façon suivante :

$$p(i) = n_i/N$$

où n_i est le nombre d'individus pour l'espèce i et N est l'effectif total (les individus de toutes les espèces).

Nous avons remarqué que certaines des métriques sélectionnées sont transformées (logarithme, pourcentage). Cela nous semble particulièrement approprié pour la Guyane ou des distributions erratiques de certaines familles ont été observées au cours de notre étude. Des taxons absents voir rares dans de nombreux cours d'eau présentaient aussi une forte abondance dans d’autres (ex: Plécoptère (Perlidae), Ephémèroptère (Euthyplociidae), Odonates (Megapodagrionidae, Perilestidae), Diptères (Dixidae, Ceratopogonidae), Coléoptère (Ptilodactylidae, Elmidae); Tichoptère (Calamoceratiidae, Ecnomidae)). Des observations similaires dans la distribution des macroinvertébrés benthiques ont également été observés dans d’autres études (Bunn et Hugues, 1997 ; Melo et Froehlich, 2001). Bunn et Hugues (1997) suggèrent que l'abondance de certains macroinvertébrés en milieu tropical est influencée par des processus issus de la réussite de la reproduction de quelques femelles. Cette stochasticité pourrait se produire sur une échelle temporelle et des espèces actuellement absents ou rares peuvent être très abondantes quelques années plus tard (ex: Plécoptère, Froehlich et Oliveira, 1997). Cela peut donc expliquer nos observations car notre zone étude se situe au niveau des têtes de bassin ou la recolonisation aérienne est considéré comme un mécanisme prédominant (Wallace, 1990).

VI.3. CALCUL DE L’IBMG

Ce paragraphe résume les différentes étapes nécessaires pour la classification d’un site/échantillon à partir des résultats ci-dessus.

VI.3.1. Avant l’échantillonnage

L’objectif principal de l’échantillonnage est défini selon le type d’opération : DCE : surveillance, opérationnel ou recherche. Une hydro-écorégion, selon les critères du tableau XV, est attribuée au site échantillonné.

VI.3.2. Récolte et préparation des données

Les invertébrés benthiques sont collectés selon la procédure établie dans le chapitre I.3.3. Les invertébrés sont identifiés au niveau familial et l’abondance des différents taxons est notée. Trois listes de taxons collectés sont établies :

- Les taxons récoltés dans les substrats organiques notés « phase A » (au nombre de 8),
- Les taxons récoltés dans les substrats minéraux notés « phase B » (au nombre de 4),
- Une liste taxonomique totale notée « AB » (au nombre de 12).

VI.3.3. Calcul l’indice

Les valeurs brutes des 6 métriques indiquées dans le tableau XVI*:* composant l’IMBG sont calculées à partir de la liste taxonomique au niveau de la famille. Les détails pour calculer les différentes métriques sont expliqués dans le paragraphe VI.2.2.

Tableau XVII: Les différentes valeurs nécessaires pour calculer l’IBMG

Reference values	**Chao1_B**	**Shannon_AB**	**CoGa.Q_AB**	**Coleo.S_AB**	**Log.Elmidae_A**	**ET.Q_AB**
HER Costal alluvial						
Sup	1.945	1.256	1.189	1.485	1.735	1.362
$moyenne_{ref}$	22.533	2.323	63.955	12.755	0.995	21.091
$écart\text{-}type_{ref}$	5.890	0.405	8.864	3.231	0.329	8.0766
HER Guiana Shield						
Sup	1.547	1.398	1.518	1.296	1.545	1.749
$moyenne_{ref}$	28.50	2.346	63.780	11.771	0.945	22.146
$écart\text{-}type_{ref}$	7.018	0.305	10.123	3.536	0.353	5.374
Inf	-3.395	-4.189	-4.130	-2.773	-2.786	-3.682

La valeur de chaque métrique est convertie en EQR :

- 1. En normalisant en fonction du type de cours d'eau (Hydro-écorégion) :

$$\text{SES} = \frac{(\text{Valeur brute} - \text{moyenne}_{ref\_HER})}{\text{écart-type}_{\text{ref\_HER}}}$$

avec 'valeur brute", la valeur mesurée de la métrique pour un point d'échantillonnage donné « moyenne$_{ref_HER}$ » et « écart-type$_{ref_HER}$ » la moyenne et l'écart-type de la distribution de la métrique en condition de référence pour le type de cours d'eau.

- 2. En calculant les écarts aux conditions de référence (EQR) par type de cours d'eau

  (bornage de 0 à 1) :

$$\text{EQRm} = \frac{(\text{SESm} - inf)}{(sup - inf)}$$

avec " SESm" : la valeur normalisée de la métrique pour un point d'échantillonnage donné ; «sup» et «inf» correspondent aux «meilleur» et «pire» valeurs pour cette métrique dans le même type de cours d'eau.
NB : Si la valeur normalisée est supérieure à la meilleure valeur, la valeur d'EQR est fixée à 1. Inversement, si la valeur est inférieure à la pire valeur, la valeur d'EQR est fixée à 0.

La note de l'IMG correspond à la moyenne pondérée des 6 métriques (EQRs) calculées précédemment en utilisant les poids (DE) indiqués dans le tableau XVI :

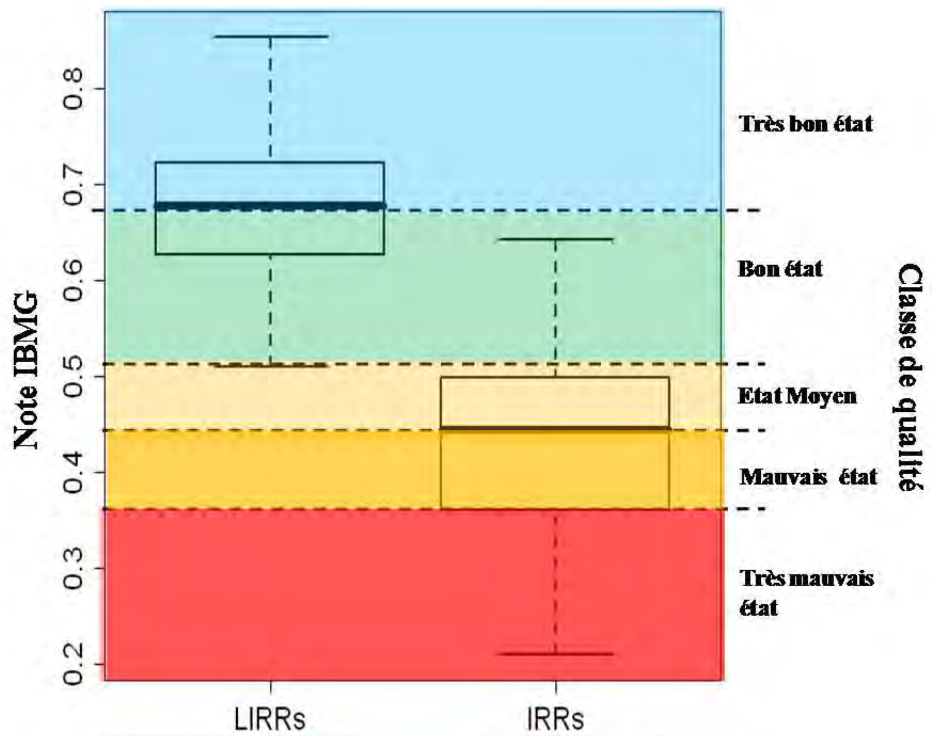
$$\text{IBMG} = \frac{\Sigma(\text{DEmoyen} \times \text{EQRmoyen})}{\Sigma\text{DEm}}$$

VI.4. LES CLASSES DE QUALITE

Comme recommandé par la DCE, l'indice doit pouvoir être interprété en termes de 5 classes de qualité écologique (« Très Bon », « Bon », « Moyen », « Mauvais » et « Très Mauvais »). Une telle interprétation nécessite la définition de limites inter-classes. Classiquement, la définition de ces classes est définie suivant la distribution des scores de l'indice sur le jeu de données de construction. Les valeurs seuils doivent être fixées de manière à ce que «la plupart des sites de référence soit de *bonne* ou *très bonne* qualité biologique» (Barbour et al., 1999).

- La moyennes des médianes des distributions des LIRRs a été pris pour limite inférieure du «Très bon état»;
- La valeur minimale de la distribution des LIRRs a été prise pour limite « Bon état/Etat médiocre » ;
- La moyennes des médianes des distributions des IRRs a été pris pour limite «Etat Moyen/Etat Mauvais»;
- Le premier quartile des IRRs a été pris pour limite «Mauvais Etat/Très mauvais état».

En appliquant ce découpage, nous avons ainsi obtenu les seuils délimitant les 5 classes de qualité (Figure 39). Ainsi, l’état écologique des stations dépend du découpage des classes de qualité. Les limites proposées ici ne sont pas arrêtées et peuvent être discutées. Elles pourront être éventuellement modifiées par l’ajout de nouvelles stations ou dans le cadre d’une inter-calibration avec les résultats obtenus sur les autres états membres (exemple : Antilles).



TME	ME	EM	BE	TBE
[0 ; 0.36]	[0.36 ; 0.42]	[0.42 ; 0.52]	[0.51 ; 0.68]	[0.68; 1]

Figure 39: Les différents seuils des classes de qualité de l’indice IBMG.

VI.5. ETAT DES LIEUX 2014 DES PME DE GUYANE SUR LA BASE DES MACROINVERTEBRES BENTHIQUES

Dans les départements d'outre-mer, l'évaluation de la qualité par la biologie est en cours de validation. Ce travail présente ici le premier état des lieux des PME de Guyane Française fait sur la base des macroinvertébrés benthiques. La faune benthique invertébrée témoigne globalement d'une **bonne qualité écologique** des PME guyanaise (Figure 40). Les résultats de l'IBMG montrent que **70%** des stations obtiennent **un bon (25%) à très bon (45%) état écologique**. Les **situations de mauvais (8%) et très mauvais Etat (7%) correspondent aux stations localisées dans les zones orpaillés.** Nos résultats ont montré que les activités aurifères au niveau des PME induisent des changements significatifs dans la diversité des macroinvertébrés benthiques. Les différences que nous avons observées sont remarquables compte tenu de l'impact localisé de ces activités dans les bassins versants. En effet, bien que nos sites d'échantillonnage soient situés en aval des zones impactées, nous avons observé des chutes drastiques de la diversité familiale (2 à 3 fois plus faible que dans les situations de référence). Si le fonctionnement écologique d'une rivière en un point donné du continuum amont-aval dépend au moins en partie des flux de matière et d'énergie provenant de l'amont, ces discontinuités propagent probablement une dégradation des fonctions écosystémiques vers l'aval. Dans le cas de l'exploitation aurifère, l'évaluation du colmatage est probablement un paramètre très important à mesurer compte tenu de l'ampleur sur le lit du cours d'eau (Figure 41). En effet, bien que certaines études d'évaluation biologique aient montré que les impacts anthropiques sont généralement uniformes pour les habitats des cours d'eau (Gerth et Herlihy, 2006 ; Lorion et Kennedy, 2009), d'autres suggèrent que les communautés de macroinvertébrés de différents substrats répondent peut-être différemment à des gradients de stress (Buss et al., 2004). Comme nous l'avons démontré dans le second chapitre, l'orpaillage influence principalement la qualité du substrat ainsi que l'augmentation des particules fines dans la colonne d'eau. Ainsi, la mise en place d'une méthode d'évaluation du colmatage et une meilleure compréhension du lien étroit taxa-habitats permettrait l'identification des sources de pressions anthropiques tout en minimisant la part des facteurs naturels non liés aux pressions dans les réponses biologiques.

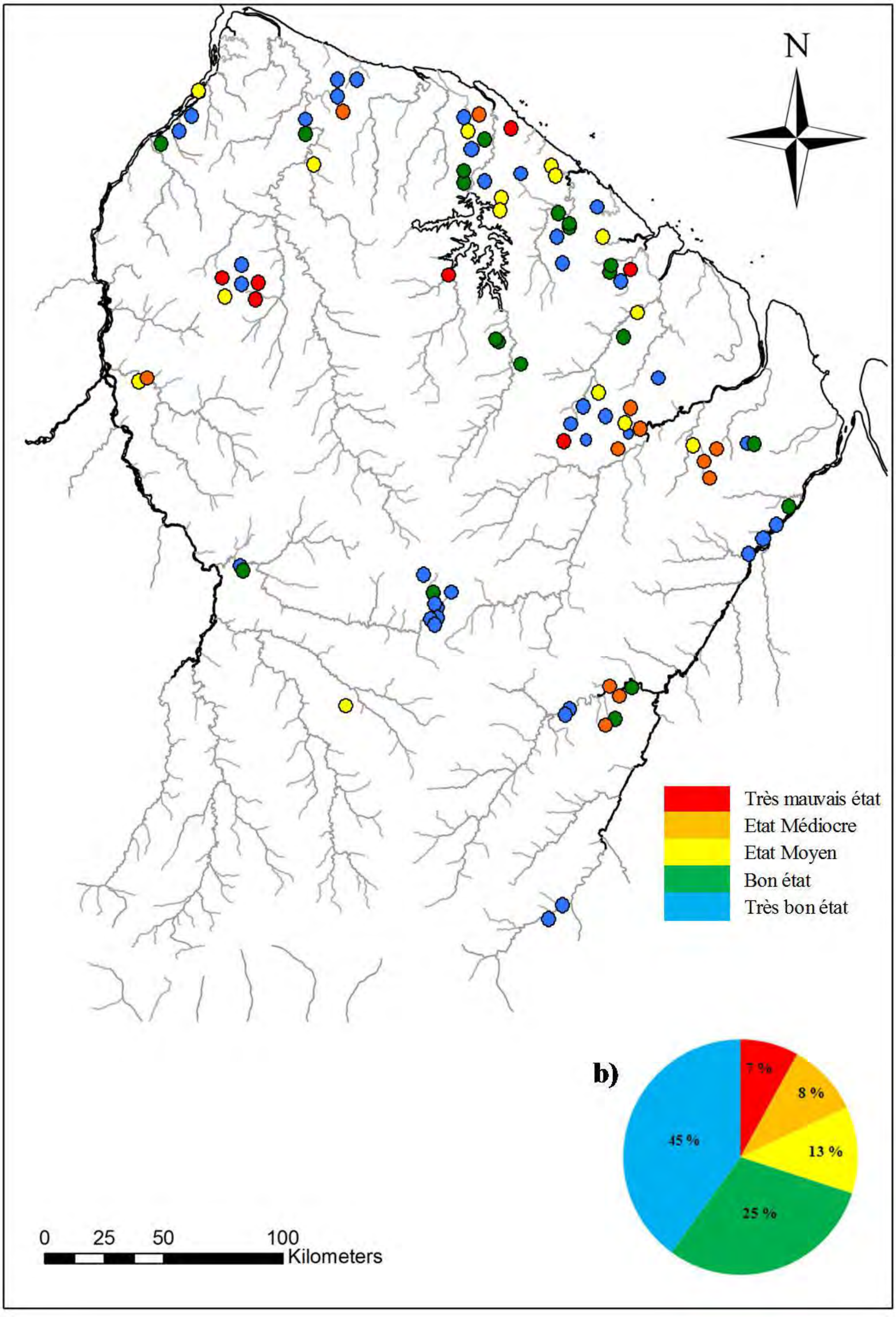


Figure 40 : a) Localisation et b) répartition des états des PME en 2014

Figure 41 : Différence d’aspect entre un cours d’eau naturel (a) et orpaillé (b). (c) Les larges quantités de sédiment colmatent rapidement le substrat

VI.6. LES LIMITES DE L'IBMG

L'indice actuel peut être utilisé en l'état pour l'application de la DCE. Parallèlement, l'IBMG pourra entrer dans un processus de calibration afin d'être affiné au cours du temps par la collection de nouvelles données situationnelles. Ce choix devra être fait avec les différents gestionnaires dans l'objectif de fixer l'IBMG final (valeurs seuils des classes de qualité). Dans l'ensemble, les notes d'IBMG valident les catégories *a priori* des sites du jeu test (Tableau XVIII).

Tableau XVIII: Résultats du jeu test : Les sites testés(Sites); l'hydro-écorégion correspondante (HER 1: Plaine alluviale, HER 2: Bouclier Guyanais); le statut a priori (Statut), le type impact observé ; la note IBMG obtenue et l'état écologique correspondant (Classe IBMG) ; et la comparaison IBMG/statut a priori (Evaluation IBMG).

Sites	HER	Statut	Type d'impact	Note IBMG	Classe IBMG	Evaluation IBMG
LaBoue1	HER 2	IRR	Orpaillage illegal	0.123	Très mauvais Etat	Bonne
LaBoue2	HER 2	IRR	Orpaillage illegal	0.259	Très mauvais Etat	Bonne
Saint-Lucien	HER 2	IRR	Orpaillage legal	0.351	Très mauvais Etat	Bonne
Orapu	HER 2	IRR	Orpaillage ancien	0.562	Bon etat	Surclassement
Tawin	HER 2	IRR	Orpaillage ancien	0.546	Bon etat	Surclassement
KapiriONF6	HER 1	IRR	Déforestation	0.452	Etat Moyen	Bonne
KapiriONF7	HER 1	IRR	Déforestation	0.53	Bon etat	Surclassement
Korossibo_2012	HER 1	IRR	Orpaillage illegal	0.546	Bon etat	Surclassement
Toussaint_2012	HER 1	IRR	Urbanisation	0.424	Etat Moyen	Bonne
RIBAL	HER 1	IRR	Carrière latérite	0.414	Mauvais Etat	Bonne
NAN3	HER 1	IRR	Carrière latérite	0.289	Très mauvais Etat	Bonne
PASS	HER 1	LIRR	Aucun	0.432	Etat Moyen	Sous-évaluation
Voltalia4	HER 2	LIRR	Aucun	0.676	Bon etat	Bonne
Kampi_2012	HER 2	LIRR	Aucun	0.507	Bon etat	Sous-évaluation
Crique-à-L'est_2012	HER 2	LIRR	Aucun	0.693	Très bon état	Bonne
Macouria_2012	HER 2	LIRR	Aucun	0.74	Bon etat	Bonne
NFS3_2012	HER 2	LIRR	Aucun	0.666	Bon etat	Bonne
Paira_2012	HER 2	LIRR	Aucun	0.68	Bon etat	Bonne
Petit_2012	HER 2	LIRR	Aucun	0.853	Très bon état	Bonne
Saul_2012	HER 2	LIRR	Aucun	0.645	Bon etat	Bonne
AFFPASS	HER 2	LIRR	Aucun	0.555	Bon etat	Bonne
SING_2012	HER 2	LIRR	Aucun	0.718	Très bon état	Bonne

Cependant, nous avons pu constater que certains sites (n=4) dont ceux anciennement orpaillés ont été «sur-classés» par l'indice (qualité écologique plus forte que celle attendue). Ces observations peuvent être dues à plusieurs raisons:

- **Un nombre de stations dans la catégorie d'impact insuffisant**. Il faut savoir que la construction de l'I_2M_2 en France Métropolitaine a bénéficié d'une large base de données nationale composée de 1725 stations pour plus de 4000 relevés (Mondy et al., 2012) alors que l'IBMG actuel a été construit à partir de 95 stations. Des travaux de calibration supplémentaires augmenteraient probablement la robustesse de l'indice et pourraient éventuellement entrainer un ré-examen des métriques (ainsi qu'une modification des classes de qualité).

- **Une ou plusieurs des metriques selectionnées répondent differemment à un type de perturbation.** En observant l'évolution de chacune des métriques indépendamment des autres (valeurs EQR propre), nous avons remarqué que la métrique relative au % d'Ephéméroptères et de Trichoptères pouvait possiblement expliquer les « sur-classements » par l'IBMG (Figure 42). En effet, ces organismes aquatiques à capacité de dispersion aérienne sont connus pour être très sensibles aux perturbations anthropiques mais aussi pour avoir une capacité de recolonisation forte (Wallace, 1990). Nous pouvons donc suspecter que ces organismes recolonisent rapidement les milieux après un impact et peuvent expliquer pourquoi les sites « anciennement perturbés » présentent des notes d'IBMG plus fortes que celles attendues.

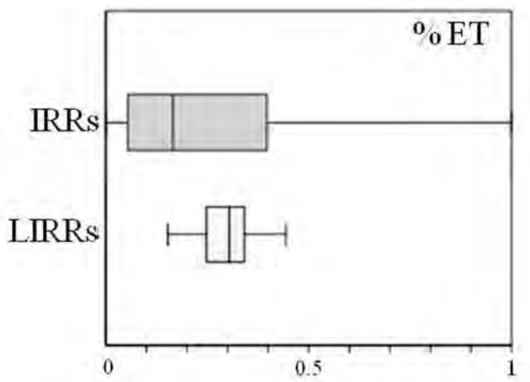


Figure 42: Valeurs d'EQR de la métrique « Pourcentage de Trichoptères et d'Ephéméroptères » (%ET) en condition de référence IRRS et impactée (IRRs) dans le jeu de donnée test.

- **La variabilité temporelle.** Lors de l'établissement des situations de référence pour un outil de bioévaluation, la prise en compte de la variabilité temporelle est un point crucial. En effet, pour assurer un suivi des sites dans des études d'impact, il est nécessaire que la référence soit toujours la même afin de mesurer l'évolution (détérioration ou amélioration) de la qualité du milieu indépendamment d'événements climatiques. Un telle problématique, impliquerait de comparer l'IBMG entre les différentes saisons sur plusieurs années. Dans notre cas, par un souci d'efficacité et de réplication, nous avons choisi d'échantillonner uniquement pendant la saison sèche. Ce choix nous a ainsi permis d'échantillonnage un nombre important de site et d'avoir une idée de l'impact en aval d'une perturbation (distance allant de 1 à 5 km selon les sites). En effet, une des questions centrales dans l'écologie des communautés porte sur la capacité de restauration et/ou cicatrisation des écosystèmes perturbés. Les états de références des PME ayant été décrit, ces informations pourront être appréhendées à l'avenir au cours des programmes de surveillance ou d'état des lieux.

VI.7. L'IBMG ET LA ZONE DE FLEUVE

Une extension aux zones aval du continuum est d'ores et déjà envisagée par les partenaires du projet. Elle devra renforcer la portée de l'IBMG, notamment par sa mise en œuvre dans les zones aval des bassins versants où se concentre la population humaine de la Guyane. L'intégration des zones aval à l'IBMG, hors eaux de transition (soumises à marées), permettra donc d'aborder la surveillance et la gestion de 95% des eaux de surface en Guyane. L'ajout certain que de nouveaux types de perturbations (ex. pollutions urbaines, petites exploitations agricoles) impliquant de nouveaux types d'impacts (ex. chimie en aval vs habitat physique en amont) devraient «favoriser» la pertinence de nouvelles métriques biologiques.

- Une première approche serait de faire un indice commun aux différentes masses d'eau. L'ajout de nouveaux de sites de référence et perturbés appartenant à un nouveau type de masse d'eau affecterait probablement la forme de l'indice actuel. Cependant, cette approche semble plus limitée car des protocoles d'échantillonnages différents ont été utilisé selon le type de masse d'eau (troubleaux (PME) vs substrat artificiel (fleuve)) et cela limiteraient par conséquence le nombre de métrique comparable (ex : pourcentage, mesures qualitatives).

- Une autre approche serait de construire un second indice spécifique aux fleuves. Cela a été le cas pour le Perou et L'Equateur (Villamarin et al., 2013) ou deux versions de l'indice IMEERA (Multimétrico del Estado Ecológico para Ríos Altoandinos) ont été développées pour chacune des hydro-écorégions ou les perturbations et le contexe biogéographique étaient différentes. Une fois les deux indices crées, une phase d'harmonisation des classes de qualité pourra être faite sur la base du principe d'intercalibration (Figure 43).

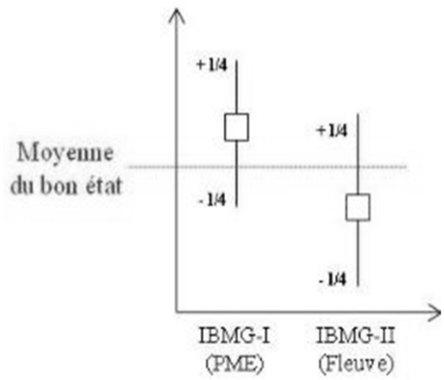


Figure 43: Exemple d'intercalibration (basé sur le principe de l'intercalibration européenne). Les carrés représentent la valeur de limite de classe de bon état et les barres verticales correspondent à l'intervalle de confiance (plus ou moins un quart de classe) de chacune de ces limites.

Conclusion générale

La bioindication en Guyane Française

En France métropolitaine, depuis que Verneaux et al., (1982) ont proposé l'indice IBGN (adapté du Trent Biotic Index, Woodiwiss, 1964), plus de trente ans de connaissance sur l'écologie de la faune benthique (Tachet et al., 2010) ainsi que de larges données issues de réseaux nationaux ont été accumulées, ce qui a permis de faire évoluer les méthodes de bioindication en accord avec les changements de politiques environnementales (Mondy et al., 2014). En ce qui concerne la Guyane Française, les problèmes inhérents à l'établissement d'un indice biologique ont été nombreux.

Le premier problème rencontré était relatif à l'état des **connaissances taxonomiques** des invertébrés de Guyane. En plus de l'outil d'évaluation biologique à mettre en place, ce travail faisait office de tout premier inventaire des Macroinvertébrés des petites masses d'eau. Au cours des deux campagnes d'échantillonnages, plus de 130.000 individus appartenant à 95 unités taxonomiques ont été collectés (Annexe I). Ce premier inventaire a permis de recenser les taxons les plus fréquent et par conséquent les plus utilisables *a priori* en bioindication. Les plécoptères, qui sont considérés comme de bons indicateurs de la qualité de l'eau dans les zones tempérées, sont rares dans les rivières des tropiques. En effet, un seul genre (*Acroneuria*) et 7 espèces ont été recensés en Amérique du Sud (Fenoglio et al., 2002 ; Fenoglio, 2007) ; et seulement une espèce a été trouvée au cours de notre étude (*Anacroneuria pictipes*, Klapálek, 1923). Nous avons aussi noté l'absence des amphipodes et des isopodes, qui sont abondants dans la zone tempérée et sont souvent utilisés comme des indicateurs pertinents de la qualité globale de l'eau (ex : *Gammarus pulex* et *Asellus aquaticus* - Whitehurst, 1991; MacNeil et al., 2002; Maltby et al., 2002). Ces derniers sont remplacés par les décapodes (crevettes, crabes) dans les cours d'eau néotropicaux, mais ont été très peu observés au cours de cette étude. Les Coléoptères, les Ephéméroptères et les Trichoptères semblent être les ordres les plus pertinents pour la bioindication en milieu tropical. Toutes ces particularités locales ont été prises en compte lors de la construction et la sélection des différentes métriques.

En milieu tempéré, de nombreuses classifications des communautés aquatiques ont depuis longtemps été établies en fonction de **leur sensibilité** à certains paramètres environnementaux, notamment chimiques tels que le pH, la salinité, les conditions trophiques, les conditions d'oxygénation (ex : Davy-Bowker et al., 2005 ; Kefford et al., 2006) ou encore la pollution organique (ex : Hilsenhoff, 1988 ; Van Dam et al., 1994). En ce qui concerne la Guyane, la sensibilité de la faune aux perturbations anthropiques a été précédemment peu étudiée. Dans le cadre de notre étude, nous avons noté que certaines familles d'éphéméroptère, de coléoptère et de trichoptère étaient particulièrement sensibles à la dégradation de type « physique » (destruction de l'habitat, augmentation de la turbidité) et étaient soit plus rares (Trichoptère (Leptoceridae), Ephemère (Polymitarcidae)) soit absentes (Trichoptères (Ecnomidae, Glossossomatidae)) dans les zones perturbées. Cependant, ces taxons n'étaient pas systématiquement présents sur les sites de référence. Ces observations ont été comfirmées au cours du Chapitre III, ou une métrique de sensibilité « CPI» (Chemical Pollution Index- Ter Braak et Prentice, 1988) a indiqué l'ubiquité de nos familles quel que soit l'impact humain sur le site (Voir valeur Annexe V). Par contre, l'étude spécifique aux éphéméroptères (chapitre IV) a mis en évidence la différenciation dans les réponses des éphéméroptères à la perturbation à un niveau taxonomique plus fin. Ces mêmes observations avaient motivé l'élaboration de l'indice SMEG basé sur ce groupe en 2001 (Thomas, 2001). Ces différentes pistes indiquent donc que les invertébrés benthiques guyanais présentent de sensibilités différentes aux perturbations humaines. Une connaissance de la faune à des niveaux taxonomiques plus fin pourrait probablement permettre de construire des métriques basées sur la sensibilité à la perturbation (Armitage et al., 1983). Dans la même optique, un indice de bioévaluation a récemment été développé pour les rivières de Nouvelle-Calédonie (Mary et Archaimbault, 2012).

Les têtes de bassin versant : un support important pour la biodiversité locale

Par leur grand nombre, les PME sont encore relativement peu touchées par les exploitations humaines et certaines présentent de haut état écologique. Cela rend la protection de ces milieux d'autant plus importante pour la conservation de la diversité régionale. Une meilleure compréhension du fonctionnement de ce type de masses d'eau permettra à terme de mettre en place une meilleure gestion. Bishop et al. (2008) soulignent que les têtes de bassin assurent des services écologiques indispensables pour l'ensemble du bassin versant et que paradoxalement nos connaissances sur le fonctionnement de ces milieux sont manquantes.

« Aqua Incognita's realm is comprised largely of the capillary network of small streams where running water begins its journey downstream to merge with other streams that contribute to rivers and lakes. We believe that these headwaters are there, but we do not know where they are, or for that matter really what they are, other than for a few small catchment outposts in the form of research sites and some pioneering synoptic surveys. »

The unknown headwaters - Bishop et al., 2008.

Cette approche des têtes de bassin en fonction des services qu'elles fournissent permet de mieux appréhender les enjeux relatifs à ces milieux peu connus. Ces auteurs présentent entre autre les services écosystémiques : d'appui (productivité primaire, cycle des nutriments, biodiversité), de régulation (régime hydrologique, qualité des eaux) et d'approvisionnement (ressource en eau, usage industriel et agricole). Comme c'est le cas en Guyane et dans d'autres endroits, les PME représentent entre 60 et 80% du linéaire total (Schumm, 1956 ; Shreve, 1969 ; Meyer et Wallace, 2001 ; Benda et al., 2005). L'un des caractères principaux que nous mettons en avant dans cette étude est que ces milieux sont probablement un **support important pour la biodiversité aquatique en Guyane**.

Malgré nos attentes de départ sur le caractère oligotrophe des eaux guyanaise (chapitre I) et la résolution taxonomique utilisée, nous avons cependant observé un fort pool d'invertébrés benthique avec 95 taxons sur l'ensemble des stations et une richesse taxonomique moyenne de 45 taxons en situations de référence. Nous avons également écarté un certain nombre de familles spécifiques à ce type de milieu (Annexe I). Cela est d'autant plus remarquable que des études précédents en Guyane (De Merona et al., 2001 ; Wasson et al., 2008) avait observé une faible richesse familiale (20 à 25 familles en moyenne). La base de donnée issue du réseau de surveillance DCE (DEAL, 2014) nous a permis de comparer trois types de masses d'eau : les PME regroupant les cours d'eau inférieur au rang 2 (notre jeu de donnée), les cours d'eaux amonts (rang 3-4) et les fleuves (supérieur au rang 5) (Figure 44). Nous remarquons que certains cours d'eaux amonts présentent aussi une forte diversité taxonomique (36 taxons en moyenne) alors que les fleuves (ordre >5) présentent des diversités taxonomiques moindres (21 taxons en moyenne). Ces observations sont à mettre en parallèle avec les études en milieu tempéré qui ont soulignée le caractère endémique de certaines espèces aux têtes de bassin (Lowe et Likens, 2005) et que le nombre d'espèces et leur effectif ont longtemps été sous-estimés (Allan et Castillo, 2007; Meyer et al., 2007).

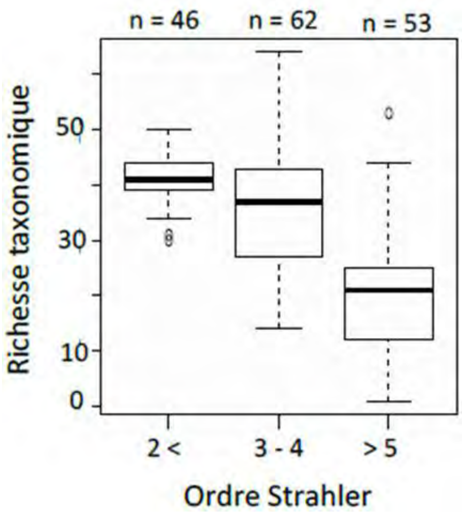


Figure 44: Comparaison des richesses taxonomiques entre les cours d'eau de différents ordres

L’approche fonctionnelle en milieu néotropical

La nature complexe des communautés naturelles et leur lien avec les activités générées par l’homme sur leur milieu rendent nécessaire le fait d'intégrer des processus agissant à différentes échelles organisationnelles. Négliger ces aspects importants des communautés, tels que la diversité fonctionnelle, peut rendre les efforts de bioévaluation moins efficace. Bonada et al. (2006) considèrent que sur les douze critères idéaux pour construire un indice basé sur les macroinvertébrés ; l’approche par les traits fonctionnels serait la plus pertinente. Par rapport à une approche taxonomique, l’utilisation des traits a l’avantage d’être prédictive, de pouvoir être reliée à des fonctions biologique et écologiques, et ne nécessite pas d’effort supplémentaire sur le terrain ou en laboratoire. L’utilisation des traits pour la bioévaluation en milieu neotropical a déjà été discutée (Tomanova et al., 2007, 2008), et certains auteurs ont proposé des indices « néotropicaux » intégrant des traits de vie (Moya et al., 2011). Cependant, sur les 22 traits bioécologiques documentés pour la faune de métropole et d’Europe (Tachet et al., 2010), seuls 5 traits (régime alimentaire, locomotion, respiration, préférence d’habitat, type de dispersion) ont pu être testés comme métriques potentielles pour le développement de l’IBMG, faute de connaissances fondamentales. Une métrique fonctionnelle a tout de même été intégrée à l’IBMG (%collector-gatherer (collecteur)). Les zones déforestées et anciennement orpaillées avaient tendance à avoir des densités plus élevées de taxons détritivores-collecteurs. Des études en milieu tropical (Wantzen et Wagner, 2006; Tomanova et al., 2006) ont néanmoins suggéré que les macroinvertébrés benthiques sont principalement omnivores en milieu tropical et posséderaient une large plasticité alimentaire selon les ressources disponibles.

Ces observations soulignent le besoin de travaux sur la quantification de nouveaux traits, notamment en relation avec la reproduction (ex : nombre de génération par an, technique de reproduction) et les cycles de vie (nombre de générations par an). Ces traits sont reconnus pour avoir un rôle important dans les capacités de résistances et de résiliences de la faune aux perturbations (Tachet et al., 2010) et ont récemment été sélectionnées pour composer l’indice multimétrique pour la France métropolitaine (fréquences relatives des espèces polyvoltines et ovovivipares – Mondy et al., 2012). Une meilleure compréhension de ces processus pourrait s’avérer particulièrement utile dans les cours eaux de tête de bassin ou la recolonisation aérienne est considérée comme un mécanisme prédominant (Wallace, 1990).

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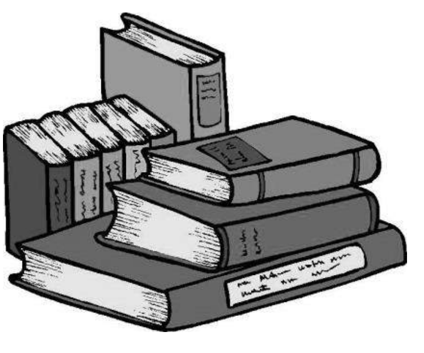

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Annexes

Annexe I: Liste des taxons rencontrés dans les petites masses d'eau de Guyane Française.

*Annexe II: «Physical habitat and water chemistry changes induced by logging and gold mining in French Guiana streams». Dedieu N., Allard L., Vigouroux R., Brosse S., Céréghino R., 2014. Knowledge and Management of Aquatic Ecosystems **415:** 02-12.*

*Annexe III: «Invertebrate communities delineate hydro-ecoregions and respond to anthropogenic disturbance in East-Amazonian streams». Dedieu N., Vigouroux R., Cerdan P., Céréghino R., 2015. Hydrobiologia **742**: 95-105.*

Annexe IV: Distribution des familles d'invertébrés au sein des quatre groupes (Annexe Article III)

Annexe V: «A multimetric macroinvertebrate index for the implementation of the European Water Framework directive in French Guiana». Dedieu N., Clavier S., Vigouroux R., Cerdan P., Céréghino R., 2015. River Research and Applications. Uncorrected Proof.

*Annexe VI: « Assessing the impact of gold mining in headwater streams of Eastern Amazonia using Ephemeroptera assemblages and biological traits». Dedieu N., Rhone M., Vigouroux R., Céréghino R., 2015. Ecological Indicators **52**: 332-340*

Annexe I : Liste des taxons rencontrés dans les petites masses d'eau de Guyane Française. Les * correspondent aux taxons spécifiques aux petites masses d'eau.

GROUP	TAXA
DIPTERA	Ceratopogonidae
	Chaoboridae
	Chironomidae
	Corethrellidae *
	Culicidae
	Dixidae *
	Dolichopodidae
	Empididae
	Limoniidae
	Psychodidae
	Simuliidae
	Stratiomyidae
	Syrphidae
	Tabanidae
	Tanypodine
	Tipulidae
EPHEMEROPTERA	Baetidae
	Caenidae
	Coryphoridae
	Ephemeridae
	Euthyplociidae
	Leptohyphidae
	Leptophlebiidae
	Polymitarcidae
ODONATA	Ashnidae
	Calopterygidae
	Coenagrionidae
	Corduliidae
	Gomphidae
	Heliocharitidae *
	Lestidae
	Libellulidae
	Megapodagrionidae
	Perilestidae *
	Plastytiscidae
	Polythoridae *
	Protoneuridae
	Pseudostigmatidae
COLEOPTERA	Carabidae *
	Dryopidae
	Dytiscidae
	Elmidae
	Gyrinidae
	Helodidae
	Hydrophilidae
	Noteridae
	Psephenidae *
	Ptilodactylidae *
	Staphylinidae
HETEROPTERA	Belostomatidae *
	Corixidae
	Gelastocoridae *
	Gerridae
	Hebridae *
	Helothrephidae
	Mesoveliidae
	Naucoridae
	Nepidae
	Notonectidae
	Veliidae
LEPIDOPTERA	Pyralidae
MEGALOPTERA	Corydalidae
	Sialidae *
PLECOPTERA	Perlidae
MOLLUSCA	Ampullariidae
	Hydrobiidae
	Hyriidae *
	Physidae *
	Planorbidae
	Sphaeriidae
	Thiaridae
CRUSTACEA/DECAPODA	Euryrhinchidae *
	Grapsidae
	Palemonidae
	Potamonidae *
	Tricodactylidae *
COLLEMBOLA	Isotomidae
DIVERS	Hirudinae
	Hydracarien
	Nematode
	Nematomorphe
	Oligochetes
	Planaria
TRICHOPTERA	Calamoceratiidae
	Ecnomidae
	Glossossomatidae *
	Helicopsychidae
	Hydrobiosidae
	Hydropsychidae
	Hydroptilidae
	Leptoceridae
	Odontoceridae
	Philopotomidae
	Polycentropodidae

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Physical habitat and water chemistry changes induced by logging and gold mining in French Guiana streams

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ABSTRACT

Key-words: *neotropical steams, headwaters, reference conditions, deforestation, gold mining*

Understanding the effects of disturbances on the physical-chemical quality of ecosystems is a crucial step to the development of ecosystem assessment tools. 95 sampling sites distributed among 4 categories of disturbance, *i.e.*: reference, logging, formerly and currently gold mining, were characterized using stream physical and chemical variables. Our hypotheses were: (i) logging and gold mining activities primarily affect the physical habitat structure of streams and (ii) both have an effect on chemical environments through nutrient and/or fine particulate resuspension. We demonstrate that physical variables describing the river bottom, and suspended solids discriminate both current and formerly gold mined sites from reference sites, while, whatever the type of impact encountered, nutrient concentrations do not prove relevant to measure human impacts. To understand distribution patterns of aquatic organism across FG, future research should thus aim at examining the match between physical-chemical and biological classifications of small streams under reference and impacted conditions.

RÉSUMÉ

Impact de l'orpaillage et de la déforestation sur la chimie de l'eau et l'habitat physique dans les petits cours d'eau de Guyane Française

Mots-clés : *cours d'eaux néotropicaux, tête de bassin, condition de référence, déforestation, orpaillage*

Comprendre l'effet des perturbations sur les paramètres physico-chimiques de qualité des eaux est une étape essentielle du développement d'outils d'évaluation. Les petits cours d'eau de Guyane Française représentent 70 % du réseau hydrographique du département, et sont soumis à de fortes pressions (déforestation et orpaillage). Nous supposons que l'exploitation aurifère et forestière affecte principalement la structure physique de l'habitat et que ces deux types d'exploitation ont une influence sur le compartiment chimique par la modification du flux de nutriments et/ou la remise en suspension de particules fines. 95 sites répartis en quatre catégories de perturbation (référence, exploitation forestière, orpaillage actuel et ancien) ont été caractérisés par des variables physico-chimiques. Nous avons démontré que les variables physiques différencient les sites orpaillés anciens et récents. Les concentrations en nutriments ne sont pas significativement modifiées par les impacts humains. Afin de mieux comprendre les patrons de distribution des organismes aquatiques de Guyane Française, le lien entre la physicochimie des cours d'eau et les communautés biologiques inféodées en situation de référence et perturbée doit être étudié.

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INTRODUCTION

Under most water management policies, ecosystem health is defined in terms of similarity to a near-pristine, undisturbed state (Bailey *et al*., 2003). In practice, predictions of the physical-chemical and/or biological conditions to be expected under undisturbed conditions in any given geographic area are based on the classification of river sites. By knowing what set of environmental conditions should be encountered at undisturbed (or least impacted) sites, one can then estimate the degree to which impacted sites are altered by human activity (Bennett *et al.*, 2011). In addition, physical-chemical classifications of rivers provide a template against which changes in biological diversity within watersheds can be interpreted in relation to natural variability and anthropogenic impacts (Van Sickle and Hughes, 2000).

Recent studies in temperate areas have prompted a large amount of characterizations of reference and impacted physical-chemical environments (*e.g.*, Tudesque *et al.*, 2008). However, differences in bioclimatic, biogeographic and geomorphological conditions preclude the transposition of current typological schemes to EU's overseas regions (see Touron-Poncet *et al.*, 2014 for a rationale), while limited scientific effort has been directed at typifying rivers in overseas Europe in terms of physical-chemical (and biological) patterns. Therefore, as a prerequisite to any methodological development, there is a pressing need to collect environmental information in a standardized manner so that fundamental data can be analyzed in an integrated way.

French Guiana (FG) is an overseas region of France located on the northern coast of South America. About 96% of its surface area (over 82 000 km^2) is covered by equatorial forest, which partly belongs to a recently-created National Park. The Guianese primary forest remains one of the least impacted of the World, however, gold mining and timber have strong impacts upon river ecosystems. Specifically, the annual gold output in the area is 60 times higher than 25 years ago (Hammond *et al.*, 2007). After the prospection of large rivers, gold industries are now focusing on smaller inland streams (Cleary, 1990; Hammond *et al.*, 2007). Small streams (from headwaters to rivers with depth $<$1 m and width $<$10 m) represent 70% of all running waters in FG. Most of small streams are located in forested areas and exhibit high ecological quality; some if not most of them have never been impacted by any human activity. In light of recent economic development, our ability to predict both reference conditions and ecosystem responses to landscape alterations will determine the success of future management actions.

We relied on extensive characterizations of stream physical (particle size, substratum heterogeneity) and chemical conditions (*e.g.*, nitrogen, phosphorus) at 95 sampling sites distributed over 95 streams and representing 4 categories of disturbance: reference (unimpacted), deforestation, ancient gold mining, and ongoing gold mining. First, we tested if impacted sites are randomly located within the river continuum or if they are characterized by particular local physical features that distinguish them from the references. Our hypothesis was that, at any given location within a stream system, gold mining and deforestation primarily affect the physical habitat structure and/or heterogeneity. Second, we tested differences in chemical variables among site categories. Assuming that both deforestation and gold mining affect chemical environments through nutrient and/or fine particulate runoff/resuspension, we expected that sediment resuspension due to gold mining would result in harsher shifts in instream environmental conditions.

MATERIAL AND METHODS

> *STUDY AREA*

This study was conducted in French Guiana (surface area = 83 534 km^2), East Amazonia, from September 2011 to December 2012. The climate is tropical moist with 3000–3400 mm of yearly precipitation mainly distributed over 280 days. There is a major drop in rainfall (dry season) between September and December and another shorter and more irregular dry period in March. The maximum monthly temperature averages 33.5 °C (32.1–35.8 °C), and the

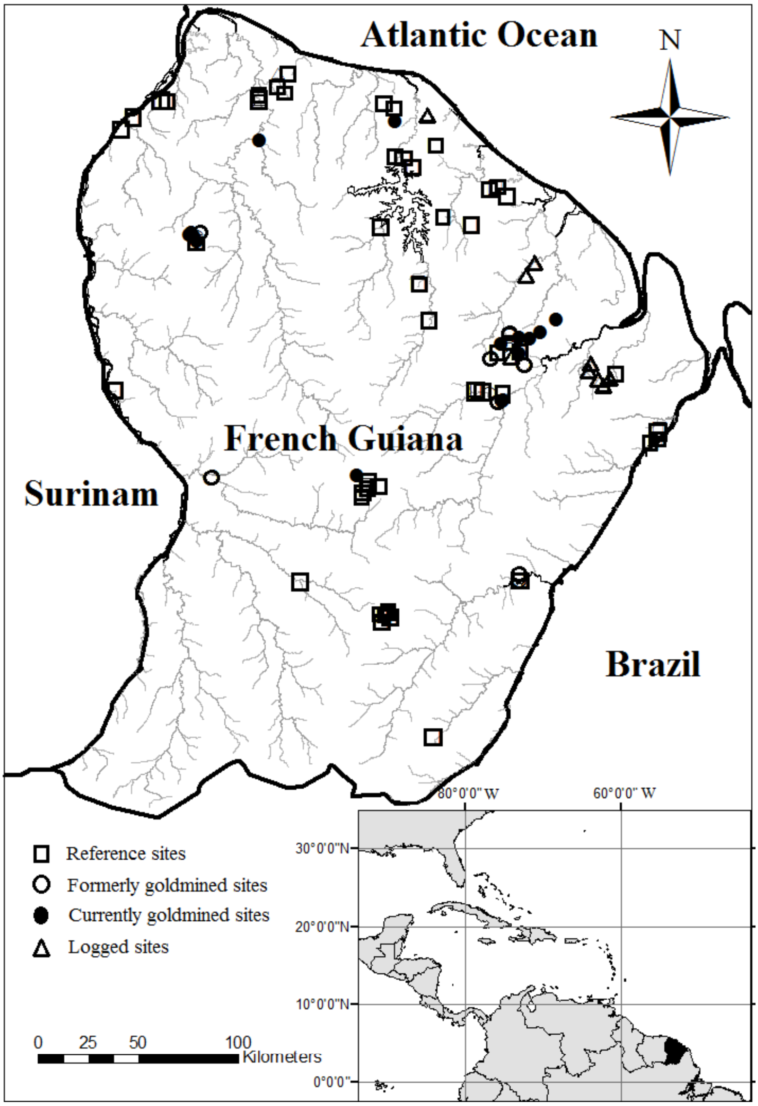

Figure 1
Map of French Guiana showing the main rivers and the location of the sampling sites.

monthly minimum averages 20.3 °C (19.7–21 °C). French Guiana's stream systems are organized around seven large rivers (Maroni, Kourou, Mana, Sinnamary, Comté, Approuagues, and Oyapock rivers); however, the "small streams" sampled in this study (water depth <1 m; stream width <10 m) represent ca. 80 000 km in total length, *i.e.* 70% of all running waters in the region. We did not considered larger streams and rivers to focus on comparable ecosystems located in the upstream part of the river continuum.

> SAMPLING SITES AND ENVIRONMENTAL VARIABLES

Our 95 sampling sites were distributed over 95 small streams belonging to FG's main river basins (Figure 1). It should be noted however that the sampling effort was higher in the Northern part of FG, due to the difficulty to access southern FG. In this specific area covered by dense rainforest and without road networks, complex logistics limited our ability to sample a larger number of sites. We thus managed to collect some samples from the main southern river basins (Figure 1). All sites were sampled during the dry season in 2011 and 2012 (September–December). Indeed, pollution is detected less efficiently during high flows because of dilution. In addition, most remote sites cannot be reached (and therefore monitored) during the rainy season. Each site was sampled once, and the sites sampled in 2012 are hence distinct from those sampled in 2011. Based on expert knowledge and field observations, sampling sites were categorized into four *a priori* groups corresponding to four types of anthropogenic pressure. Reference sites (Ref, $n = 57$) were defined as sites not subjected to anthropogenic impacts such as gold mining, deforestation, chemical pollution, agricultural or urban runoff. Deforested sites were subjected to logging for wood products and timber (Log, $n = 15$), under the supervision of the National Forest Office (ONF). The ONF manages a sustainable logging industry based on strict plans intended to minimize impacts on the environment. The remaining sites were formerly subjected to gold mining but no longer exploited (Fog, abandoned mining $n = 9$), or currently subject to gold mining (Cug, $n = 14$). Either formerly or currently gold mined sites refer to illegal gold mining activity. "Illegal" mining refers here to so-called "informal" mining, *i.e.*, small-scale traditional (or artisanal) mining which also occurs in most South American countries (Hammond *et al.*, 2007). Mercury is used for gold amalgamation during the mining process, and about 30% of the mercury is released into the river. Mercury concentration in the water column is however insignificant (see Coquery *et al*., 2003; Maury-Brachet *et al*., 2006 for documented cases in FG). In addition, the effects of heavy metals on the composition of biological communities expected to form Biological Quality Elements in subsequent developments are not obvious (De Jonge *et al*., 2008). Although mercury was not taken into account in our study, it should be noted however that river sediments have a strong adsorption capacity for heavy metals (Pfeiffer *et al.*, 1989; Roulet *et al.*, 1999) and may certainly be a relevant parameter to quantify the impact of gold mining. Mercury can subsequently accumulate in plant and animal tissues before entering food chains – in French Guiana some concentrations in edible parts of locally consumed fish can surpass the advisory level for human consumption, thus forming a key concern for human health (Durrieu *et al*., 2005).

Stream scale variables, namely elevation above sea level, distance from the source, and slope were obtained from Geographic Information System (GIS, ESRI ArcGis 10). These variables characterise the location of sampling sites within the upstream-downstream river continuum. Site scale variables were chosen to describe the heterogeneity of riverbed substrate and habitat availability at each site. They were recorded directly in the field and accounted for the percentage composition of organic and mineral substrate types, using the standardized protocol by Souchon *et al*. (2000). These variables included: %leaf litter (Litt), %submerged roots on the banks (Bank), %submerged vegetation, mostly macrophytes (Macr), %woody debris (Wood), %Silt, %Sand (particle size $<$2 mm), %Gravel (Grav 2–25 mm), %coarse substratum (Coar $>$ 25 mm). Coarse substrates being scarce in French Guiana, this category of mineral substrate included pebbles, boulders, and/or rocky outcrops. In addition, we recorded the stream width (Widt, m) and water depth (Dept, m). The forest canopy coverage (Fore) was evaluated visually, from 0 to 100% (Table I). We also measured chemical variables accounting for the chemical impairment of stream ecosystem (PO_4^{3-} and NO_3^-) by human activities and for the transport of solids (Total Suspended Solid and Turbidity) (Table II). Turbidity was measured directly in the field using an Eutech Instruments Turbidimeter (TN-100). Other chemical analyses were carried out at Hydreco Laboratory, Petit-Saut, based on water samples taken at each site and immediately frozen. Chemical analyses followed standard methods summarized in AFNOR (2000–2005).

> *DATA ANALYSIS*

We first used a Principal Component Analysis (PCA) to ordinate the sites according to topological and physical variables, and to bring out potential shifts in physical conditions following anthropogenic disturbance. Prior to analysis, continuous variables were log-transformed, and discrete variables expressed in percentages were Arcsin-transformed. Plots of the first two ordination axes usually capture most of the variance and consequently contain most of the information that is likely to be interpretable (Waite *et al.*, 2000). Neighbouring sites

Table I

Environmental variables used to describe the four streams category (Ref : reference; Log; logging; Fog: formerly gold mined; Cug: current gold mined). Values indicate mean ± SD.

	code	unit	Ref	Log	Fog	Cug
Stream scale variables						
Elevation	Elev	m a.s.l.	131.3 ± 153.7	57.2 ± 18.1	80.1 ± 36.3	90.8 ± 24.9
Distance from headwater source	Dist	km	3.3 ± 3.8	1.9 ± 2.4	3.6 ± 2.6	5.61 ± 3.4
Slope	Slop	‰	4.7 ± 3.6	4.7 ± 3.4	4.7 ± 3.19	3.84 ± 2.6
Site scale variables						
Bank	Bank	%	12.1 ± 13.2	14.1 ± 12.1	9.2 ± 7.2	7.79 ± 5.4
Macrophyte	Macr	%	3.1 ± 17.4	3.7 ± 13.7	2.2 ± 6.5	0.83 ± 2.7
Litter	Litt	%	24.1 ± 19.8	24.1 ± 23.9	16.7 ± 19.8	25.5 ± 31.3
Woody debris	Wood	%	14.1 ± 12.9	20.9 ± 22.3	13.2 ± 11.6	7.26 ± 7.9
Silt	Silt	%	11.9 ± 18.1	16.3 ± 17.5	21.7 ± 12.3	26.2 ± 27.7
Sand	Sand	%	47.2 ± 31.9	49.5 ± 24.7	26 ± 28.17	16.4 ± 20.4
Gravel	Grav	%	20.7 ± 22.9	11.4 ± 15.4	35.7 ± 27.8	30.7 ± 31.6
Coarse substratum	Coar	%	19.6 ± 25.9	12.7 ± 15.4	26.6 ± 25.5	26.7 ± 27.8
Width	Widt	cm	379.9 ± 231.5	429.3 ± 271.2	393.6 ± 198.7	394.3 ± 187.5
Depth	Dept	cm	25.5 ± 11.9	26.7 ± 14.2	23.1 ± 7.9	27.9 ± 13.3
Forest coverage	Fore	%	74.8 ± 16.3	55.1 ± 28.3	57 ± 27.4	64.6 ± 23.6

Table II

Chemical variables used to assess human disturbance on the four streams category (Ref: Reference; Log: logging; Fog: formerly gold mined; Cug: current gold mined). Values indicate mean ± SD.

Chemical variables	unit	Ref	Log	Fog	Cug
Turbidity	NTU	5.31 ± 22.5	5.51 ± 11.1	32.5 ± 93.4	27.6 ± 43.9
NO_3^-	mg/L	0.31 ± 0.16	0.25 ± 0.21	0.34 ± 0.08	0.25 ± 0.15
PO_4^{3-}	mg/L	0.04 ± 0.02	0.04 ± 0.05	0.02 ± 0.02	0.02 ± 0.02
Total suspended solid	mg/L	6.1 ± 12.8	8.4 ± 12.9	9.9 ± 14.4	60.4 ± 155.4

in the scatterplots were expected to define areas with similar physical environments. Conversely, sites having a large distance to each other were expected to be distant in the feature space, according to environmental characterization. In order to compare distributions of sites according to disturbance types, a Kruskal-Wallis (KW) test was performed on the site coordinates of the two first axes of the PCA. To further bring out relationships between water chemistry, local environments and disturbance, significant differences among *a priori* groups were also tested using Kruskal-Wallis tests on the raw values of measured parameters. Then, significant differences in physical characteristic between *a priori* groups of sites (Ref, Log, Fog, Cug) were further assessed using Wilcoxon tests.

As different sites were sampled in 2011 and 2012, the sampling year was not informative and we hence pooled the two years data. All computations were performed using the R Software (R Development Core Team, 2003), the ADE-4 (Thioulouse *et al.*, 1997) and Vegan (Oksanen *et al.,* 2013) package.

RESULTS

Eigenvalues for axis 1 and 2 of the PCA were 3.43 and 2.26, respectively (Figure 2a). The first and second axes explained 24.53% and 16.7% of the overall variance, respectively. The distribution of sampling sites in the scatterplot did not show clear clumps according to environmental characteristics, but rather displayed a predictable, upstream to downstream gradient. Axis 1 thus displayed a gradient of elevation, slope and substratum size (from high (left) to low (right)). These parameters are related to the river competence (*i.e.* the maximum

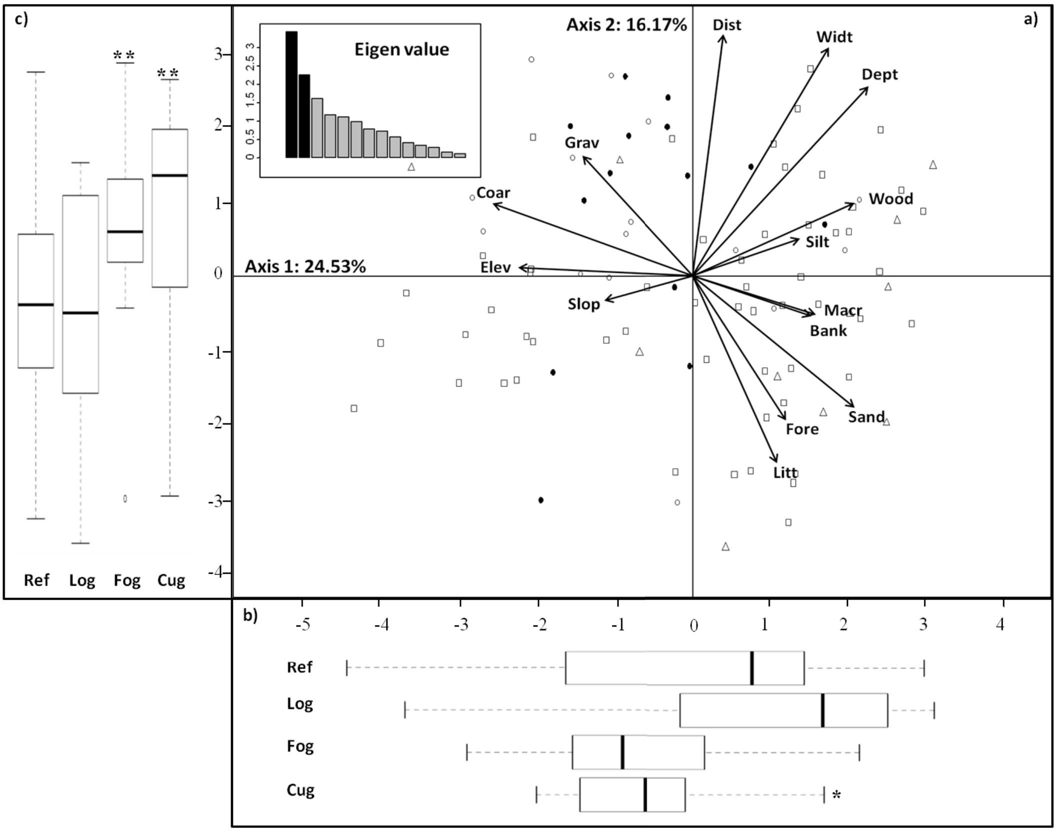

Figure 2
(a) Principal Component Analysis (PCA) biplot showing the distribution of the 95 sites according to environmental variables (see table I for acronyms). Rectangles: reference sites; triangles: logged sites; open circles: formerly gold mined sites and filled circles: currently gold mined sites. (b) Boxplots of the coordinates of the sites on the first axis. (c) Boxplots of coordinates of the sites on the second axis. Ref: reference sites; Log: logged sites; Fog: formerly gold mined sites; Cug: currently gold mined sites. (: p-value $<$ 0.1; ***: p-value $<$ 0.01).*

size or weight of material a river can transport). Axis 2 accounted for stream width and depth, and distance from source (from high (upper area) to low (down)).

Sites subjected to current and former gold mining were mostly distributed along axis 2, while sites subjected to logging were distributed along axis 1. Only reference sites and sites subjected to current gold mining differed significantly in their distribution along axis 1 (Figure 2b), currently exploited sites being more concentrated in the upstream areas. When the distribution of sites was examined along axis 2 (Figure 2c), both formerly and currently gold mined sites differed from other sites, while reference sites and sites subjected to logging did not show significantly different distributions.

Stream scale variables showed significant differences between impairment categories. The mined sites had coarser substrates than the references and logged sites, as shown by a significant difference in the percentage composition of mineral particles. Significant differences were found in %sand (KW-chi-square = 15.3776, *df* = 3, *p*-value = 0.001521), %silt (KW-chi-square =12.2781, *df* = 3, *p*-value = 0.006489) and %gravel (KW-chi-square = 7.6945, *df* = 3, *p*-value = 0.04277). Such a difference between reference and mined sites holds true for both the formerly and currently gold mined sites.

Considering chemical variables, neither PO_4^{3-} and NO_3^- (Figures 3 and 3b), nor suspended solids and turbidity (Figures 3c and 3d) showed significant differences between reference sites and sites subjected to logging. Gold mining did not alter PO_4^{3-} and NO_3^- concentrations (Figures 3a and 3b), however, stream turbidity values (Figure 3d) were significantly different

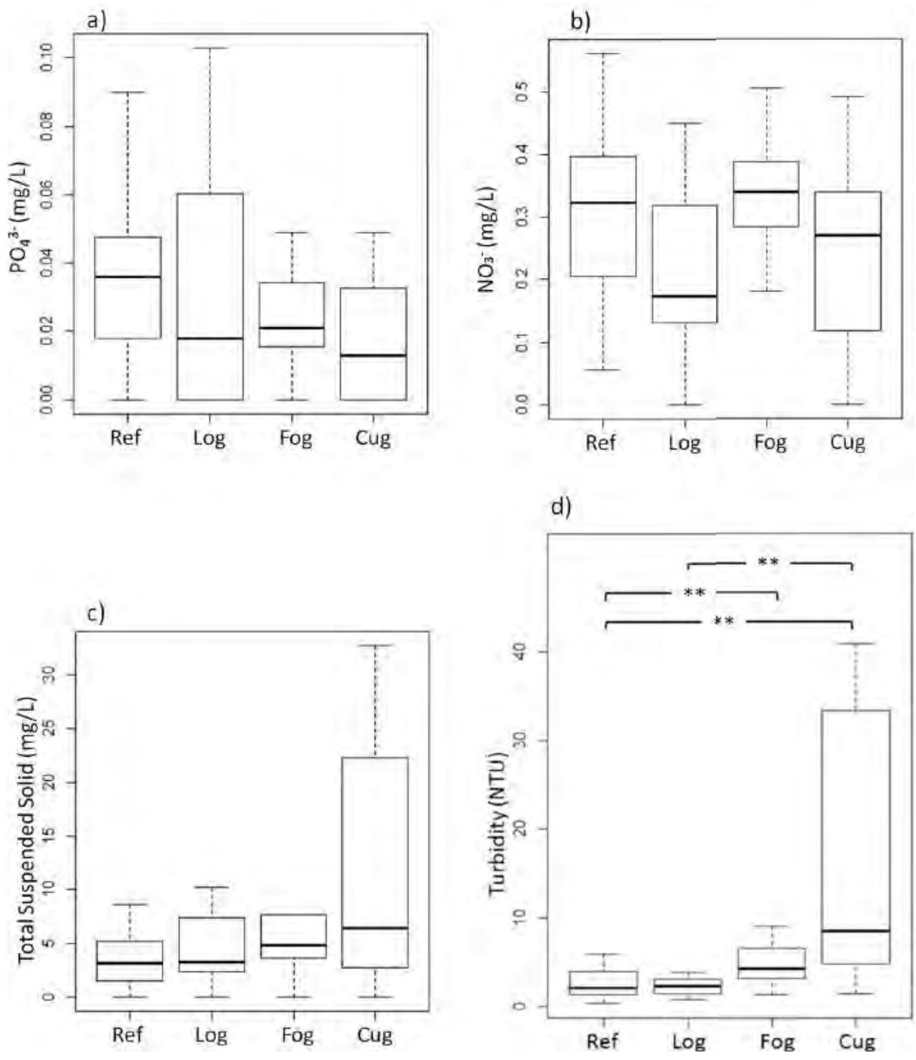

Figure 3
*Boxplots of chemical variables (a) PO_4^{3-} (mg/L), b) NO_3^- (mg/L), c) Total Suspended Solid (mg/L) and d) turbidity (NTU)) in reference (Ref), logged (Log), formerly goldmined (Fog) and currently goldmined (Cug) sites. Limits of the box represent the first and third quartiles, bold line is the median, and whiskers are extreme values. Stars indicate the significance of Kruskal Wallis test between classes (**: p-value $<$ 0.05).*

from values observed at reference and logged sites (see appendix I for outputs of Wilcoxon tests).

DISCUSSION

Given the contrasted types of human activities that affect stream ecosystems in FG, we expected significant differences in physical-chemical characteristics of impaired streams in relation to disturbance type.

Sites subjected to logging (deforestation) had finer bottom substrates than the reference sites. This can be related to increased inputs and deposits of fine particles, brought to the stream through tractor tracks and gravel road creation for logging trucks (Forman and Alexander, 1998). Such a tendency is triggered under equatorial climate, where harsh rains (rainy season) have a strong erosive effect on bare lands (Dudgeon, 2008). Despite increased siltation, suspended solids and water turbidity were not affected, highlighting the moderate effect of logging on streams in FG. It should be noted however that logging is strictly controlled in FG, in order to minimise environmental impacts. Specific measures include the absence of clear-cutting, and the protection of the riparian zone where logging is forbidden. In the same way, logging trucks cannot be used during the rainy season and cannot cross streambeds, thus reducing sediment load to the aquatic ecosystems (Panchout, 2010). Such management efforts seem to prove efficient, as we did not detect any significant effect on turbidity and other chemical variables that are usually strongly sensitive to intensive logging activities. Specifically, leaching of soils and canopy opening are known to modify nitrogen and phosphorus fluxes to the aquatic ecosystem (Sweeney *et al.*, 2004; Neill *et al.*, 2006).

The river bottom of most gold mined sites was characterised by a dominance of gravels. This trend is not related to natural processes. Gold deposits are collected by washing the soils adjacent to the streams with high pressure water jets, and gravels are then sieved and released to the stream (Hinton *et al.*, 2003), therefore increasing their prevalence over the streambed. During these operations, the streams also receive the draining water that contains a high load of sediments (Watts *et al*., 2003), explaining the higher turbidity at gold mined sites (see also Mol and Ouboter, 2004; Brosse *et al.*, 2011).

Contrary to our expectation that gold mining, through the predicted clearing of the riparian forest and soil leaching, should increase stream eutrophication (see Hammond *et al*., 2007; Palmer *et al.*, 2010), we did not find any significant change in nutrient loads (PO_4^{3-} and NO_3^{-}) in gold mined sites. This result can be explained by the illegal nature of the exploited sites, which typically remain hidden under the canopy and do not host more than 30–40 workers (Hinton *et al.*, 2003). There is therefore no deforestation at these sites, and hence no drastic shift in nutrient fluxes. This however does not mean that the ecosystem is not impaired. In particular, it has been demonstrated that both fish and diatom assemblages are strongly affected by the turbidity generated by small scale gold mining through habitat clogging and changes in light penetration over the bottom (Cleary, 1990; Mol and Ouboter, 2004; Brosse *et al*., 2011; Tudesque *et al.*, 2012).

It is worth noting that turbidity remained significantly higher in formerly gold mined sites than in reference sites. This is probably due to fine sediment storage in the stream pools, so that these sediments can be re-suspended in the river column when river discharge increase during the frequent rain events. Therefore, temporary physical disturbances of stream ecosystems can persist in time, explaining why biological assemblages do not recover after stopping mining (*e.g.*, fish and diatom, see Brosse *et al*., 2011; Tudesque *et al.*, 2012).

Finally, the comparison of formerly and currently gold mined sites revealed a shift of activities towards the upstream sites. This might be afforded to two non-mutually exclusive reasons. First, the rarefaction of the gold resources as well as the rise of gold prices brought gold miners to move deeper in the forest (Cleary 1990; Hammond *et al*., 2007) and exploit more remote upstream sites. Second, the increased control of illegal gold mining by French authorities (Coppel *et al.*, 1998) forces illegal miners to exploit those remote sites and to remain as inconspicuous as possible. This probably explains why in most of our sites we did not observe deforestation that would make the mining sites easily detected (Hinton *et al.*, 2003).

In conclusion, we demonstrated that, under unimpacted conditions, there is no clear clustering of freshwater streams in French Guiana, thus complicating aims to set up a stream typology based on physical characteristics. Also, whatever the type of impact encountered in small streams of FG, nutrient concentrations did not prove relevant to measure human impacts. Logging did not result in detectable impacts on stream physical characteristics, probably because this type of activity is strictly managed and controlled by local stakeholders. However, site scale variables that describe the riverbed, habitat and suspended solids (*i.e.* simple physical measurements) clearly segregated both currently and formerly gold mined sites from reference sites. These results highlight the persisting, adverse effect of mining on the benthic habitat. Assuming that the structure of biological communities in streams are not due to random processes (Minshall and Petersen, 1985) but is strongly influenced by physical factors such as stream bed morphology (Wallace and Webster, 1996), hydrological conditions (Power *et al.*, 1988), one can assume that substrate homogenization by anthropogenic activities will largely constraint the benthic community structure. Moreover, invertebrates or diatoms are tightly integrated into the structure and functioning of the benthic ecosystem, one may expect dramatic decreases in the biological quality of headwater streams with the shift of gold mining towards the upstream areas. To design potential biological indication tools of impairment, future research should thus aim at examining the match between physical-chemical and biological classifications of small streams under reference and impacted conditions.

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Appendix I: Wilcoxon tests results. Bold values correspond to significant difference between the two tested categories. Ref: reference sites, Log: logged sites, Fog: formerly gold mined sites, Cug: currently gold mined sites.

Axe1	Reference sites	Logged sites	Formely gold mined sites	Currently gold mined sites
Ref	–	*W* = 213, *p*-value = **0.1257**	*W* = 57, *p*-value = 0.1057	*W* = 506, *p*-value = 0.07218
Log	–		*W* = 127, *p*-value = **0.0204**	*W* = 120, *p*-value = **0.0159**
Fog	–			*W* = 98, *p*-value = 0.78
Cug	–			
Axe2	**Reference sites**	**Logged sites**	**Formely gold mined sites**	**Currently gold mined sites**
Ref	–	*W* = 318, *p*-value = 0.7963	*W* = 222, *p*-value = **0.006539**	*W* = 210, *p*-value = **0.0092**
Log	–		*W* = 49, *p*-value = 0.08289	*W* = 44, *p*-value = **0.007**
Fog	–			*W* = 93, *p*-value = 0.062
Cug	–			

%Silt	Reference sites	Logged sites	Formely gold mined sites	Currently gold mined sites
Ref	–	*W* = 144.5, *p*-value = **0.006095**	*W* = 399.5, *p*-value = 0.8559	*W* = 217.5, p-value = **0.01171**
Log	–		*W* = 124, *p*-value = **0.03197**	*W* = 65, *p*-value = 0.08254
Fog	–			*W* = 87.5, *p*-value = 0.5833
Cug	–			
%Sand	**Reference sites**	**Logged sites**	**Formely gold mined sites**	**Currently gold mined sites**
Ref	–	*W* = 285, *p*-value = 0.7697	*W* = 568.5, *p*-value = **0.02584**	*W* = 604, *p*-value = **0.001086**
Log	–		*W* = 123.5, *p*-value = **0.03527**	*W* = 129, *p*-value = 0.3008
Fog	–			*W* = 129, *p*-value = **0.004485**
Cug	–			
%Gravel	**Reference sites**	**Logged sites**	**Formely gold mined sites**	**Currently gold mined sites**
Ref	–	*W* = 371.5, *p*-value = 0.2322	*W* = 266.5, *p*-value = **0.03589**	*W* = 317, *p*-value = 0.3091
Log	–		*W* = 35, *p*-value = **0.01409**	*W* = 112, *p*-value = 0.6666
Fog	–			*W* = 48, *p*-value = 0.1143
Cug	–			

Turbidity	Reference sites	Logged sites	Formely gold mined sites	Currently gold mined sites
Ref	–	*W* = 164, *p*-value = 0.713	*W* = 164, *p*-value = **0.043**	*W* = 217.5, *p*-value = **0.006095**
Log	–		*W* = 134, *p*-value = 0.64	*W* = 115, *p*-value = **0.00015**
Fog	–			*W* = 341, *p*-value = 0.6833
Cug	–			

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Annexe III

PRIMARY RESEARCH PAPER

Invertebrate communities delineate hydro-ecoregions and respond to anthropogenic disturbance in East-Amazonian streams

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Abstract Many tropical regions lack models predicting the biological and environmental conditions expected in any given area, thus precluding the implementation of reference condition-based water policies. We focused on streams of French Guiana, and tested two predictions: geomorphology determines ecological sub-regions that have typical invertebrate communities, and diversity declines as anthropogenic pressure increases. Sixty-five stream sites were sampled for benthic invertebrates and physical–chemical variables across various watersheds. We used the Self-Organizing Map algorithm (neural network) to model relationships between invertebrate communities and environmental variables. Sites characterized by invertebrate communities clustered into two major subsets matching French Guiana's hydro-ecoregions: the coastal alluvial plain characterized by recent sediment and low elevations, and the Guiana Shield characterized by an eroded rocky substrate and dense rainforests. Changes in community composition, and to a lesser extent taxonomic richness within each sub-region revealed ecological impacts of gold mining and logging, further clustering hydro-ecoregions into subsets of reference and impaired sites. Further analyses would, however, be needed to identify tipping points between natural and disturbed states, especially in remote headwater streams where gold mining had the harsher impact upon freshwater diversity, making upstream communities resembling the most downstream impacted ones.

Keywords Benthos · Neotropical rivers · Reference conditions · Self-organizing maps · Water Framework Directive

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Introduction

Intended to reach a "good ecological status" of all water bodies by 2015, Europe's Water Framework Directive (WFD) has prompted a large amount of works which yielded characterizations of either reference physical–chemical environments and biological communities in continental Europe, as well as practical tools (e.g., biological indices) to evaluate water quality (Borja, 2005). Overseas regions of Europe occur in various biogeographic areas of the World (Atlantic, Caribbean, Pacific, Indian Oceans). These regions have the same water policy objectives as the continental ones, but they were overlooked during recent developments of bioassessment tools that fulfill the WFD guidelines. Differences in bioclimatic, biogeographic, and geomorphological conditions, as well as differences in anthropogenic pressure preclude the transposition of typological schemes and bioassessment tools developed in continental Europe to overseas regions. For instance, biological traits (life history patterns, body size, etc.), species richness, and numerical dominance do not compare among biogeographic regions. Last but not least, the development of bioassessment methods in many overseas regions suffer from a lack of taxonomic knowledge, so that ecologists are faced with minimal background on the distribution patterns of aquatic species. For instance, very little is known about macroinvertebrate taxonomy and distribution in headwater streams of French Guiana, East-Amazonia.

French Guiana (FG) is an overseas region of France located on the north-eastern coast of South America. About 96% of its surface area (83,534 km^2) is covered by a remarkably species-rich equatorial forest (Bongers et al., 2001). The Guianese primary forest remains one of the least impacted of the World, however, gold mining and timber have strong localized impacts upon river ecosystems. Specifically, the annual gold output in the area is 60 times higher than 25 years ago (Hammond et al., 2007). After the prospection of large rivers, gold industries are now focusing on smaller inland streams (Cleary, 1990; Hammond et al., 2007). Small streams (from headwaters to rivers with depth <1 m and width <10 m) represent 70–80% of all running waters in FG. Most small streams are located in forested areas and exhibit high ecological quality; some if not most of them have never been impacted by any human activity. In light of recent economic development, there is a pressing need to identify reference (undisturbed) conditions that will then allow environmental managers to estimate the degree to which human activities have altered stream ecosystems.

While providing new basic information on freshwater diversity and its environmental drivers in eastern Amazonia, this study takes a step toward the implementation of the WFD in French Guiana (one of France's 11 inhabited overseas regions) by bringing out the first classification of FG streams. Routine surveys conducted by local consultancies revealed changes in river communities in relation to local anthropogenic pressure (Vigouroux et al., 2005). However, because FG is mostly covered by dense (inhabited) rainforest deprived of road networks, the local to regional distribution patterns of macroinvertebrates are fundamentally unknown, especially in the remote, headwater streams. By using ordination and classification of 65 sampling sites distributed throughout FG, we tested the following predictions: (i) geomorphology determines ecological sub-regions that have typical macroinvertebrate assemblages and species richness, and (ii) invertebrate diversity broadly declines as anthropogenic pressures increases. Environmental explanatory variables were used to interpret invertebrate diversity and distribution, and the resulting schemes were discussed in the context of water policy.

Materials and methods

Study area

This study was conducted in French Guiana, from September 2011 to December 2012. The climate is wet tropical with 3,000–3,400 mm of annual precipitation mainly distributed over 280 days. There is less rainfall (dry season) between September and December and another shorter and more irregular dry period in March. The average monthly maximum temperature is 33.5°C (32.1–35.8°C), and the average monthly minimum is 20.3°C (19.7–21°C). French Guiana's streams flow into seven large river watersheds (Maroni, Kourou, Mana, Sinnamary, Comté, Approuagues, and Oyapock rivers). It is worth noting that "small streams" (water depth <1 m; stream width <10 m) represent ca. 80,000 km in total length, i.e., 70–80% of all running waters in the region.

Sampling sites and environmental variables

We sampled 65 sites belonging to different watersheds distributed throughout the country (Fig. 1). The sampling effort was inevitably higher in the northern range, due to the limited access to the south. The

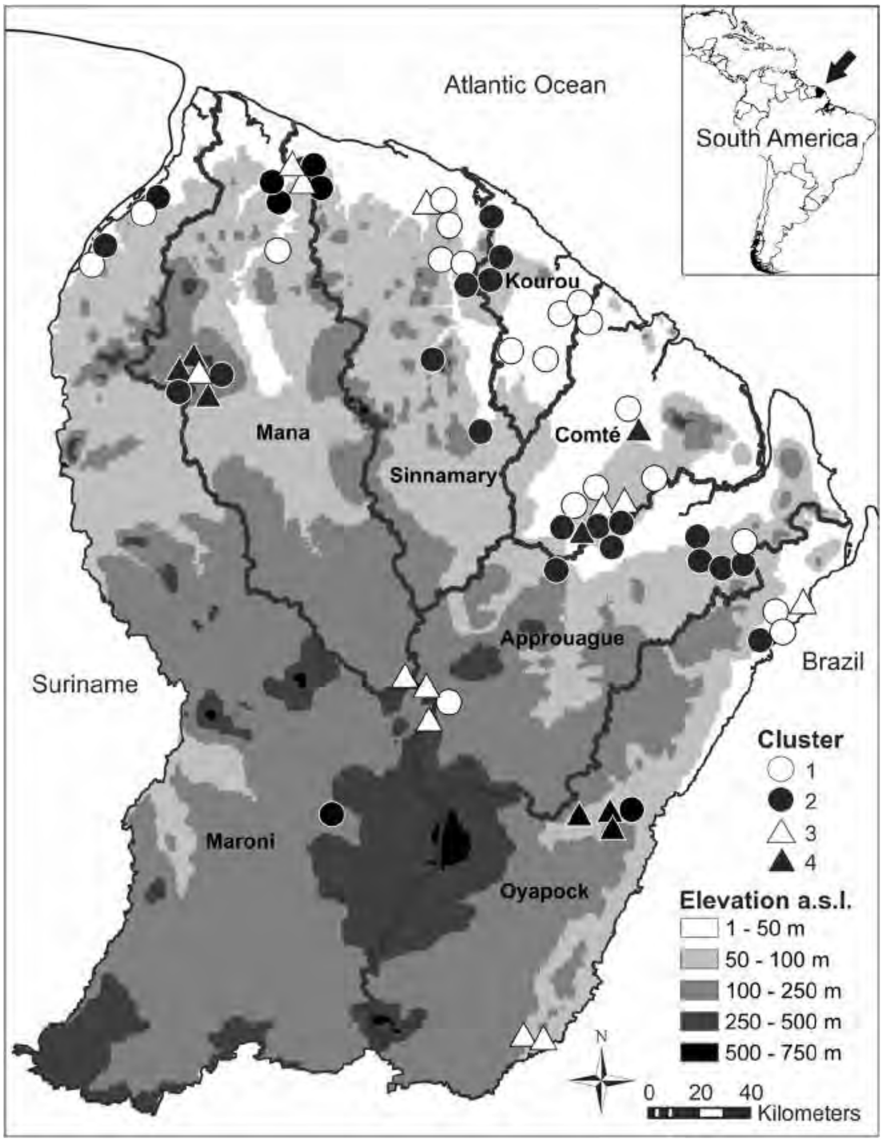

Fig. 1 Distribution of the 65 sampling sites in French Guiana. Different markers are used to assign sites to clusters 1–4 depicted in Fig. 2

complex logistics needed to obtain samples from this remote range limited the number of sites. We nevertheless managed to sample southern sites from the main river basins (Fig. 1). All sites were sampled during the dry season (September–December) in 2011 and 2012. Impacted sites were subjected to two major disturbance types: gold mining ($n = 11$; either legal or illegal), and land-use impacts ($n = 14$; logging for wood products and timber, runoff from small cultivations and/or urban areas). Reference sites ($n = 40$) were defined as sites not subjected to anthropogenic impacts.

All sampling sites were characterized using topological, morphological, water chemistry, and habitats variables (Table 1). For each site, a Geographic Information System (GIS, ESRI ArcGis 10) was used to obtain elevation above sea level (m a.s.l.), distance from the source (km), and slope (per mil). These variables were chosen because they characterize the location of sites within the upstream–downstream river continuum. Water samples for chemical analyses were taken at each site between 10:00 a.m. and 2:00 p.m. (to minimize potential diurnal variation in the data) and immediately frozen. Chemical analyses

Table 1 Main physical–chemical characteristics of the 65 sampling sites in French Guiana

	Mean ± SE	Minimum–maximum
Topology		
Distance from source (km)	3.52 ± 3.49	0.5–16
Elevation (m.a.s.l)	171.6 ± 78.04	32.46–234.64
Slope (per mil)	3.66 ± 2.79	0.34–11.58
Mean stream width (m)	4.06 ± 2.27	1.04–13.56
Mean stream depth (cm)	27.4 ± 11.81	5.25–55.06
Substrate composition		
% Silt	15.54 ± 17.92	0–90
% Sand	45.21 ± 31.55	0–100
% Gravel	20.37 ± 20.79	0–85
% Coarse substratum	18.93 ± 24.95	0–85
% Woody debris	12.94 ± 10.4	0–41.6
% Macrophytes	10.56 ± 16.46	0–70
% Litter	21.92 ± 19.89	0–85
% Roots on the bank	12 ± 10.53	0–50
Water chemistry		
pH	5.73 ± 0.79	4.04–7.65
Temperature (°C)	24.96 ± 1.53	19.9–30.7
Conductivity (μs cm^{-1})	32.62 ± 24.69	12–135
Dissolved oxygen (mg l^{-1})	6.37 ± 1.45	3.7–7.9
Turbidity (NTU)	12.37 ± 44.87	0.51–329
Total suspended matter (mg l^{-1})	16.3 ± 70.07	0–566
Dissolved organic carbon (mg l^{-1})	18.15 ± 14.38	0–78
Nitrate (μg l^{-1})	0.29 ± 0.15	0–0.83
Total phosphorus (μg l^{-1})	0.04 ± 0.04	0–0.172

were carried out at Hydreco Laboratory (Petit-Saut, French Guiana) following standardized methods (AFNOR 2000, 2005a, b). Chemical variables measured in the laboratory were: turbidity (NTU), total suspended matter (mg l^{-1}), NO_3 (μg l^{-1}), Total Phosphorus (μg l^{-1})), and Dissolved Organic Carbon (mg l^{-1}). Four variables were directly measured in the field using probes: % oxygen (WTW 3205®), turbidity (EUTECH®), pH (WTW 3110®), and conductivity (WTW 3110®). Water temperature (°C) was the mean of values given by all above-mentioned probes.

The length of a site was defined as 10 times its width, and transects were established each 5 meter along this length, for subsequent habitat measures. Water depth (m) and the percentage composition of organic and mineral substrate types were determined on a 1 m^2 area every meter along each transect. Mean water depth at a site was the mean of all point measurements. Stream width (m) was the mean value of all transects. The substrate types included: % litter, % submerged roots on the banks, % macrophytes, % woody debris, % silt, % sand (particle size <2 mm), % gravel (2–25 mm), and % coarse substratum (>25 mm). Coarse substrates being scarce in FG streams, pebbles, boulders, and rocky outcrops were grouped in a single category.

Macroinvertebrates sampling

Twelve sample units were taken at each site, i.e., 8 samples in organic substrates (roots, macrophytes, aquatic plants, litter, and bryophytes) and 4 samples in mineral substrates (pebbles, gravels, and sand), thus representing the average distribution of these substrate types in FG streams. Sample units in organic substrates consisted in intensive sweeping of a hand net (frame size = 46 × 23 cm; mesh size = 500 μm) during 1 min over a 0.46 × 1.5 m^2 area (net width × 1.5 m^2). Sample units in mineral substrates were obtained by dragging a 5-cm layer of sediment with the same net, over a 0.46 × 1.5 m^2 area. Prior to dragging, coarse particulates (pebbles) were brushed in front of the net, and then removed. The samples were preserved in the field in 4% formalin (final concentration). Invertebrates were sorted in the laboratory and preserved in 70% ethanol. They were mostly identified to family and enumerated (list of taxa and mean numbers of individuals per m^2 in electronic supplementary material).

Data analysis

To sort the 65 sampling sites according to the invertebrate communities, we used the Self-Organizing Map algorithm (SOM Toolbox version 2 for Matlab®, see Vesanto et al. (1999) for practical instructions). The strengths of the SOM in comparison with conventional multivariate analyses were discussed in Giraudel & Lek (2001). Briefly, combining ordination and gradient analysis functions, the SOM is convenient to visualize high-dimensional data in a readily interpretable manner without prior transformation. Here, it is worth noting that conventional (multivariate) ordination and classification techniques were inefficient at revealing patterns of community organization, certainly because we had to analyse organism counts with skewed distributions (due to many zero values). The SOM algorithm is an unsupervised learning procedure that transforms multidimensional input data into a two-dimensional map subject to a topological (neighborhood preserving) constraint (Kohonen, 2001). The SOM, thus, plots the similarities of the data by grouping similar data items together onto a 2D-space (visualized as a grid) using an iterative learning process (Park et al., 2003). The SOM algorithm is specifically relevant for analyzing sets of variables that vary and co-vary in non-linear fashions, and/or that have skewed distributions. Additionally, the SOM algorithm averages the input dataset using weight vectors through the learning process and thus removes noise. A full description of the modeling procedure employed here (training, map size selection, number of iterations, map quality measurements) was detailed in Céréghino & Park (2009).

The structure of the SOM for this analysis consisted of two layers of neurons connected by weights (or connection intensities): the input layer was composed of 86 neurons (one per invertebrate taxon) connected to the 65 sampling sites, and the output layer was composed of 42 neurons visualized as hexagonal cells organized on an array with 7 rows and 6 columns. The number of 42 output neurons was retained after testing quantization and topographic errors (see Céréghino & Park, 2009). At the end of the training, each site is set in a hexagon of the SOM map. Sites appearing distant in the modeling space (according to invertebrate data used during the training) represent expected biological differences for real environmental characteristics.

Ward's algorithm was applied to cluster the trained map (Ultsch, 1993). The SOM units (hexagons) were divided into clusters according to the weight vectors of the neurons, and clusters were justified according to the lowest Davis Bouldin Index, i.e., for a solution with low variance within clusters and high variance between clusters (Negnevitsky, 2002).

In order to analyze the contribution of each invertebrate taxon to cluster structures of the trained SOM, each input variable calculated during the training process was visualized in each neuron (hexagon) of the trained SOM in gray scale. This visualization method directly describes the discriminatory powers of input variables (here invertebrates) in mapping (Kohonen, 2001), while allowing to bring out invertebrate distribution patterns. To investigate relationships between physical–chemical and biological variables, we introduced the 22 physical–chemical variables into the SOM previously trained with the abundance data for the 86 invertebrate taxa (see Céréghino & Park, 2009). During the training, we used a mask function to give a null weight to the 22 physical–chemical variables, whereas biological variables were given a weight of 1 so that the ordination process was based on the 86 invertebrate taxa only (Compin & Céréghino, 2007). Setting mask value to zero for a given component removes the effect of that component on organization (Sirola et al., 2004).

Model structures (clusters of sites) were visualized using GIS. Significant differences in taxa richness, evenness (Simpson index), and entropy (Shannon–Weaver index) among SOM clusters were tested using Kruskal–Wallis tests.

Results

After training the SOM with the invertebrate densities at 65 sites, the sites were classified into four subsets (clusters 1–4) according to the quantitative structure of their macroinvertebrate communities (Fig. 2a). Clusters were plotted on a geographical map of FG in order to ease interpretations (Fig. 1). Sites in clusters 1–2 and 3–4 corresponded to two major geographic areas of FG, i.e., coastal areas and inland forests, respectively. Within coastal ranges, sites subjected to logging (TETE, KAP 1, KAP 2, KAP 4 and KAP 5) and gold mining (KORO, ROSE, BORD, GREN, BOIB) were grouped in cluster 2 in the left-bottom area of the map. All sites subjected to agricultural or urban runoff (LUCI, REL1, REL2, DACH, PLAM, SM, APA, BAST, and HUMU) belonged to coastal areas, but did not show clear grouping. Within inland forest ranges, sites submitted to gold mining (QUAD, CLAN, CHS2, and CHAF) were grouped in cluster 4 in the left-top area of the map. Therefore, based on site status (reference *vs* impacted) each major geographic area was further separated into two sub-groups of sites according to a gradient of anthropogenic impact ranging from low (right area of the SOM, clusters 1 and 3) to high (left area, clusters 2 and 4).

Only 9 taxa out of 86 (e.g., Noteridae, Sialidae, Lestidae) occurred in one specific cluster of sites (see

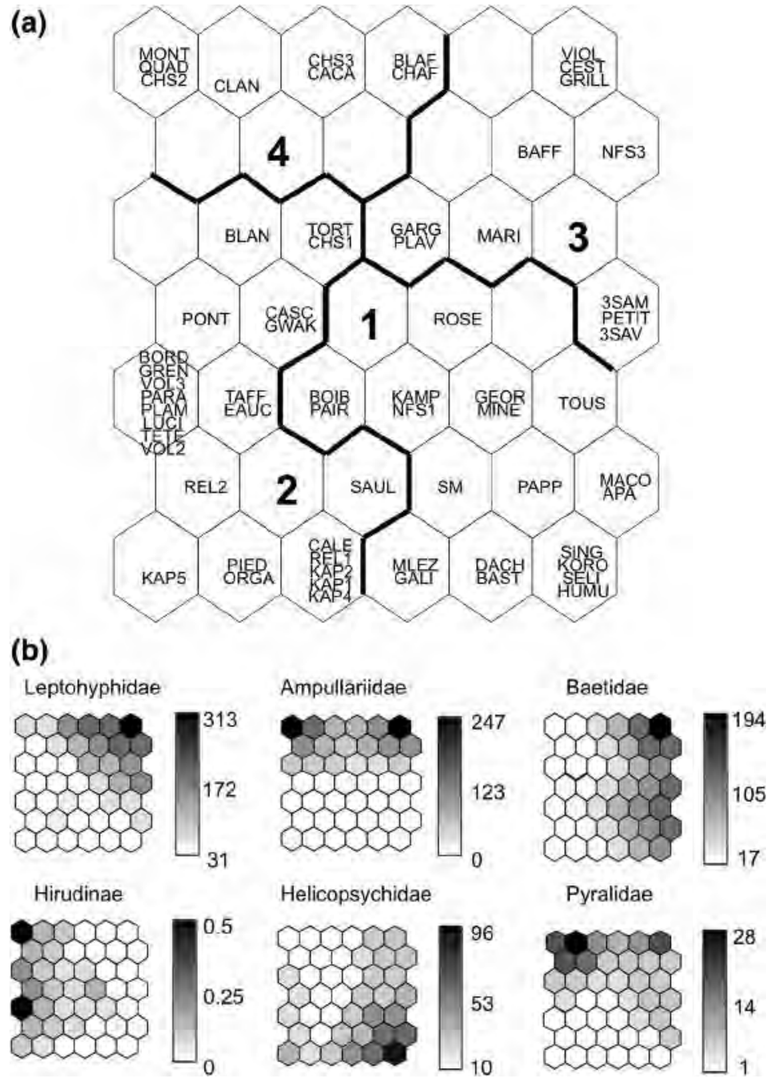

Fig. 2 **a** Distribution and clustering of the 65 sites on the self-organizing map (SOM) according to the abundance of 86 macroinvertebrate taxa. Codes within each hexagon (e.g., MONT, QUAD) correspond to sites. Clusters 1 to 4 were derived from Ward's algorithm. **b** Gradient analysis of density (number of individuals per m^2) for a few selected taxa on the trained SOM represented by a shaded scale (*dark* high density, *light* low density). Each small map representing taxa that follow similar patterns can be compared to the map representing the distribution of sites in (**a**), thus showing the distribution patterns of the various taxa (in shades of *gray*) within each sub-area of the SOM

electronic supplementary material). When the distribution of each taxa was visualized on the trained SOM using a shading scale (examples in Fig. 2b), Baetidae and Caenidae (Ephemeroptera), Notonectidae (Heteroptera), Limoniidae (Diptera), and Polycentropodidae (Trichoptera) were characteristic of unimpacted sites, whatever the geographic area (clusters 1 and 3). Hirudinae, Oligochaeta, and Nematodes were frequent in impacted sites (clusters 2 and 4). Higher densities for these taxa, therefore, indicated anthropogenic impacts, rather than regional differences in stream habitat conditions. Sites in forest ranges (clusters 3-4) showed higher densities (and occurrences) for invertebrate families belonging to the *Mollusca* (Ampullariidae, Hydrobiidae, and Thiariidae) (see examples in Fig. 2b). Such taxa, therefore, had strong influence upon the classification. Cluster 3 (forest, reference sites) showed higher densities for Leptohyphidae and Leptophlebiidae (Ephemeroptera), Megapodagrionidae and Calopterygidae (Odonata), Ceratopogonidae, Empididae, Culicidae and Simuliidae (Diptera), Elmidae (Coleoptera), and Planaria. Cluster 4 was characterized by higher densities for Ephemeroptera (Euthyplociidae), Plecoptera (Perlidae), Dryopidae (Coleoptera), Lepidoptera (Pyralidae), Psychodidae (Diptera), and Odonata (Plastytiscidae). Sites in coastal ranges (clusters 1 and 2) had a lower number of typical taxa, namely Coryphoridae (Ephemeroptera), Euryrhinchidae, and Helicopsychiidae (Trichoptera). Sites in the cluster 1 were characterized by

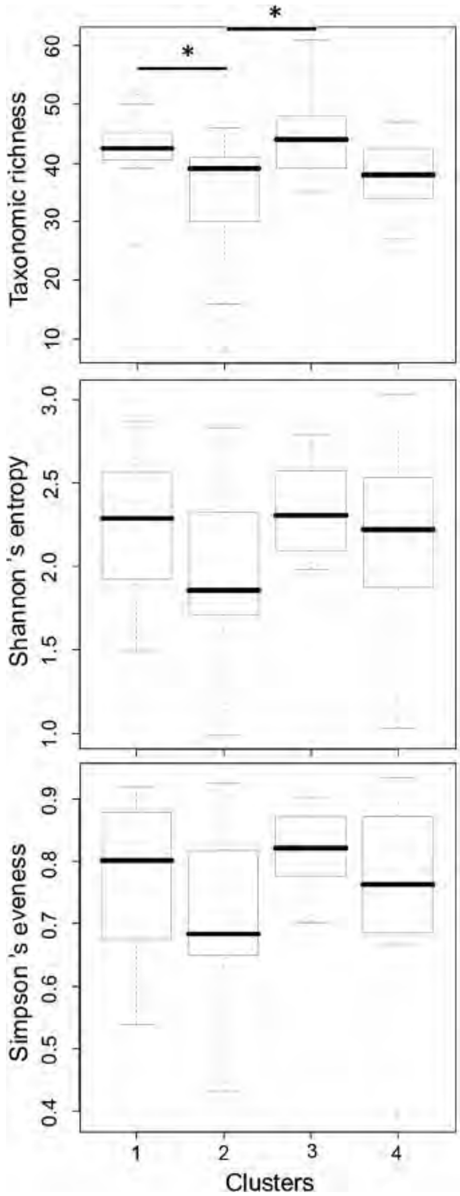

Fig. 3 Boxplots of diversity metrics distributions (taxonomic richness, Shannon's Entropy, Simpson's evenness), in the four clusters derived from the SOM clustering. Significant differences between groups or clusters were tested with Kruskal–Wallis tests; *Asterisk* significant differences at $P < 0.05$

higher densities of Helicopsychidae, Glossosomatidae and Odontoceridae, (Trichoptera), Corethrellidae (Diptera), Coryphoridae, and Polymitarcyidae (Ephemeroptera). Sites in cluster 2 were characterized by low numbers of taxa and individuals. Finally, the comparison of community diversity indices between clusters only showed significant differences for taxonomic richness: the number of invertebrate taxa was significantly lower in cluster 2 than in cluster 1 ($P = 0.0015$) and cluster 3 ($P = 0.015$, Fig. 3).

When environmental variables were introduced into the SOM previously trained with abundance data for 86 invertebrate taxa at the 65 sites (Fig. 4), sites in clusters 1–2 were characterized by finer substrates (sand and silt) as well as higher %macrophytes, %litter, and %dead wood. Sites in clusters 3–4 had higher values for elevation, conductivity, pH, and %coarse mineral substrates. Specifically, physical–chemical variables that indicate human impacts (higher values for turbidity and suspended matters from right to left areas of the map) confirmed the gradient of disturbance within each sub-region, from cluster 1 to 2, and 3 to 4.

Discussion

This study provides new information on the environmental determinants of freshwater invertebrate diversity and taxa distribution in eastern Amazonia, while proposing the very first biological typology of FG streams. Previously, Chandesris & Wasson (2005) suggested an abiotic typology of FG watersheds by delineating hydro-ecoregions based on geomorphological, hydrological, and climate data. Delineated according to macroinvertebrate communities, clusters 1 and 2 in our study match the "*coastal alluvial plain*" characterized by recent sediment and low elevations, while clusters 3 and 4 correspond to the inland "*Guiana Shield*" characterized by an eroded rocky substrate, a variability of elevations and large stream systems under a dense forest coverage. These results also indicate that in the streams of FG, the structure of macroinvertebrate communities changes along a longitudinal gradient, from inland headwaters to the coastal rivers. First of all, sites in cluster 3–4 had coarse bed paving substrates, and hosted richer and more diverse invertebrate communities than the sandy sites of cluster 1–2. Downstream changes in tropical stream invertebrate communities were previously related to changes in ecological processes along gradients of elevation (i.e., from up- to down-stream areas), notably changes in leaf litter inputs and algal production (Sites et al., 2003). In FG, however, there is no such clear gradient of elevation above sea level.

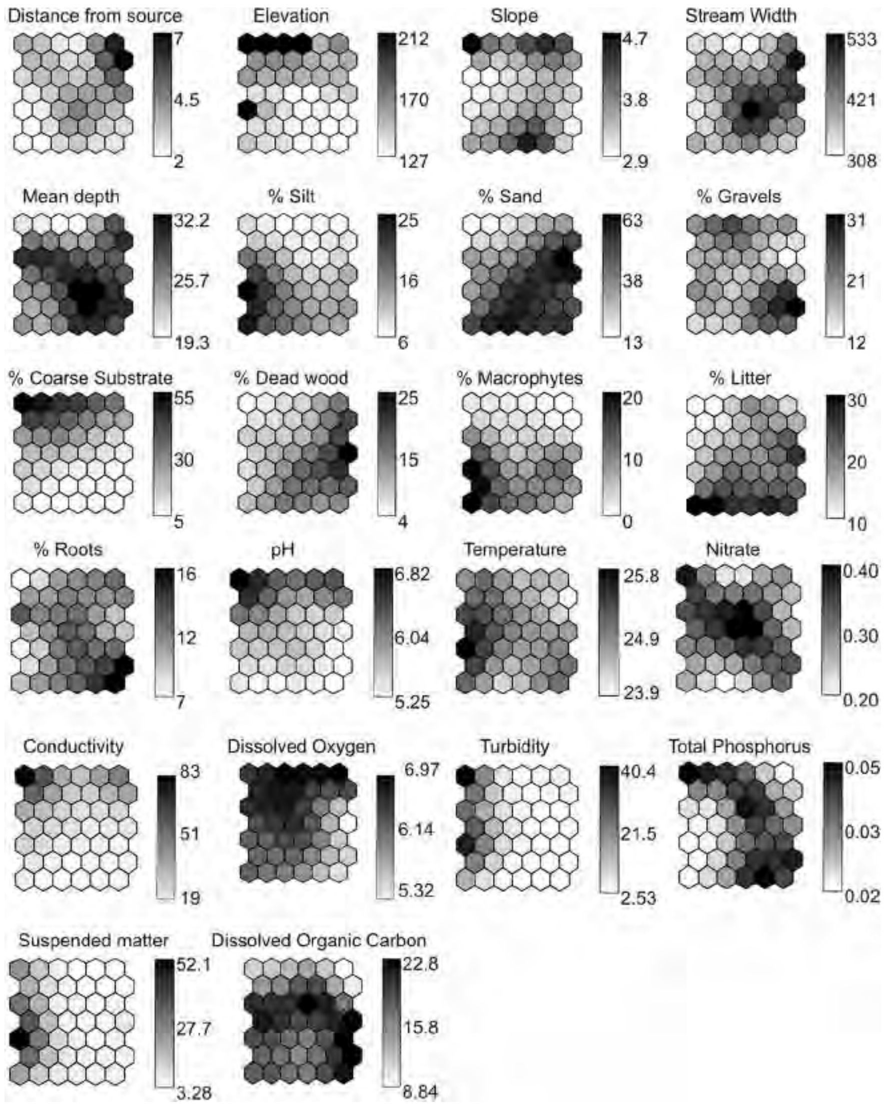

Fig. 4 Gradients of selected environmental variables on the SOM previously trained with macroinvertebrate data. The mean value for each variable was calculated in each output neuron of the SOM. Dark represents a high value, while light is a low value. See Table 1 for units

Inland forest ranges must be seen as a dense "sea of hills" forming higher elevation islands within a low-elevation matrix. Hence, on a local scale, steep slopes alternate with long flat plateaus. Because river competence determines substrate size and, subsequently, invertebrate diversity (Buss et al., 2004; Arrington & Winemiller, 2006; Salman et al., 2013), some inland sites close to the headwater sources can group together with coastal sites within the framework of a biological typology (e.g., sites KAMP CALE and NFS1).

There was also a clear difference between clusters of sites in terms of pH. Coastal sites correspond to acidic waters, locally called "black waters" because of their darker color. pH values in headstream waters were neutral. Only a few invertebrate families were strictly characteristic of neutral waters, however, suggesting that most taxa have broad pH tolerance. Those taxa specific of neutral streams belong to Mollusca and Ephemeroptera. The formation of the shell of freshwater mollusca notably requires neutral to basic pH (Merritt & Cummins, 1996). The sensitivity of Ephemeroptera to acidification has been previously demonstrated in temperate rivers (Dangles et al., 2004; Petrin et al., 2007) but not in tropical areas yet. Insect families like Euryrhinchhidae, Corydalidae, Odontoceridae Helicopsychidae, and the Ephemeroptera Polymitarcidae are mainly found at low pH in FG streams, meaning that they tolerate acidity. However, because of their life style, these taxa also have a strict preference for a given substrate type (Merritt & Cummins, 1996). Odontoceridae and Helicopsychidae caddisflies require sand to build their larval case, and the Polymitarcidae is a specialist burrower in silt and sandy substrates. Coryalidae are mainly observed under dead wood. Such striking, selected examples support the hypothesis that in naturally acid stream, community composition does not only depend on water acidity but also on substrate size.

Ordination and cluster analyses are frequently used in the exploratory phase of typologies. All sites were included in our SOM, regardless of a priori consideration of disturbance. By doing so, we expected that geographically adjacent sites appearing distant in modeling space (according to macroinvertebrate communities) would represent differences among sites in biological quality. Sites subjected to anthropogenic disturbance grouped into specific clusters within large hydro-ecoregions, suggesting that disturbance has an effect on freshwater diversity but did not override geomorphological controls of the distribution of macroinvertebrates in FG streams. Gradients of disturbance were apparent both within coastal plains and forest ranges, revealing ecological impacts of gold mining and logging. Previous studies demonstrated that bank erosion due to these activities increase sediment upload, to the detriment of freshwater diversity (Cleary, 1990; Mol & Ouboter, 2004; Hammond et al., 2007). In addition, gold miners extract and crush coarse mineral substrate, further homogenizing river beds and generating high turbidity that decrease light penetration into the water to the detriment of epilithic algae, an important base of the food chains for invertebrate (Sloane-Richey et al., 1981; Graham, 1990). Interestingly, a few impacted sites located in the inland forest, namely TAFF, BORD, TORT, and GREN, were assigned to cluster 2 (impacted sites, coastal area) instead of cluster 4 as we could have expected. These sites are typically subjected to small-scale illegal mining. Illegal settlements are increasingly located in remote areas, and are not cleared by gold miners in order to remain invisible to aerial and satellite surveillance (Hammond et al., 2007; Coppel et al., 1998). While former illegal gold mining concentrated on downstream river reaches, there is little information on the impact of this new trend of gold mining activities (Mol & Ouboter, 2004; Mendiola, 2008; Yule et al., 2010; Brosse et al., 2011). Our analyses, therefore, suggest that gold mining in headwater, forested streams may have the harsher impact upon freshwater diversity because it is likely to generate strong longitudinal discontinuities, making upstream communities resembling the most downstream impacted ones.

All sites subjected to less severe disturbance (runoff from small cultivations and/or urban areas) were located in the coastal alluvial plain (clusters 1 and 2), which concentrates 80% of FG's human population. The distribution of these sites did not follow a clear gradient of disturbance within the corresponding hydro-ecoregion, suggesting that small-scale increases of dissolved organic carbon and/or nitrogen and/or phosphorus concentrations that typically match agricultural and/or urban activities were not large enough to generate significant deviation from predictable communities. Given that nutrient inputs are limited to diffuse runoff from small cultivations and/or sparse habitations, we assume that, in the absence of gold mining and timber, the proportions of fine mineral substrate and organic substrates (submerged roots, macrophytes, dead wood, litter) play a key role in determining invertebrate diversity at these coastal sites.

While Europe's WFD provided compelling reasons for developing river typologies, reference schemes, and pressure-impact models in member States, the lack of published study for overseas regions first reflects minimal knowledge of the distribution patterns of aquatic species in "neotropical Europe", especially in remote headwaters (but see recent works by Bernadet et al. 2013 and Touron-Poncet et al. 2013 in the Carribean). Our analysis revealed that invertebrate communities show qualitative and quantitative spatial patterns, but also change in terms of biological traits in relation to natural conditions and anthropogenic disturbance (e.g., increasing mollusk diversity in forest sites, insects at reference sites, annelids at impacted sites). Hence, our typology will prove useful in defining impacted and least impacted river reaches for the upcoming development of a WFD-compliant biological index for FG (Mondy et al., 2012). Our study, however, shows that human pressure have an impact on FG streams but no clear gradient of disturbance was observed, i.e., our impacted sites were subjected either to negligible (diffuse runoff) or harsh disturbance (gold mining, logging). In other words, intermediate disturbance is clearly lacking in FG. Assuming that modern biological indices must be scaled against a gradient of water quality corresponding to different levels of impairment (typically representing “high”, “good”, “moderate”, “poor” and “bad” ecological quality), we may anticipate difficulties in defining intermediate quality classes between “bad” and “good” quality. Hence, further analyses of physical–chemical environments would be needed to identify tipping points between natural and disturbed states.

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Annexe IV : Distribution des familles d'invertébrés au sein des quatre groupes. Les nombres indiquent les abondances moyennes (individus moyens par $m^2 \pm$ écart type) (Chapitre III).

Phylum	Class / Order	Family / Subfamily	Cluster 1	Cluster 2	Cluster 3	Cluster 4
Nematoda			-	-	0.08±0.11	0.59±0.98
Nematomorpha			0.07±0.13	0.46±0.23	-	0.26±0.12
Planaria			7.76±4.93	0.91±0.52	4.93±15.31	2.67±1.39
Hydracarina			1.08±1.16	0.56±0.73	3.19±3.30	1.31±1.80
Annelida	Hirudinae		0.01±0.03	0.09±0.04	-	0.06±0.04
	Oligochaeta		5.66±4.81	13.39±12.34	1.56±1.58	5.94±.25
Mollusca	Gastropoda	Ampullariidae	-	-	8.86±23.83	13.68±24.53
		Hydrobiidae	0.56±1.44	0.25±2.20	11.23±29.71	4.74±7.80
		Planorbidae	-	0.39±18.56	0.03±0.06	0.33±0.22
		Sphaeriidae	-	0.15±0.62	0.57±1.10	0.44±1.14
		Thiaridae	-	-	0.83±2.12	3.15±7.94
	Decapoda	Euryrhinchidae	0.01±0.04	0.08±0.18	-	-
		Palaemonidae	1.77±1.21	0.99±0.99	1.75±2.27	1.36±1.10
		Potamonidae	-	0.01±0.02	-	0.06±0.12
		Tricodactylidae	-	-	0.04±0.11	0.30±0.49
Insecta	Coleoptera	Dryopidae	0.11±0.22	-	0.27±0.42	0.33±0.39
		Dysticidae	0.99±1.10	0.12±0.24	2.31±3.01	0.07±0.12
		Elmidae	11.56±11.22	4.86±3.85	52.47±74.56	8.32±5.25
		Gyrinidae	0.17±0.22	0.08±0.15	0.09±0.1	0.06±0.12
		Helodidae	1.48±1.78	0.34±0.52	2.32±1.28	0.50±0.85
		Hydrophilidae	0.73±0.83	0.14±0.30	1.33±1.58	0.19±0.34
		Noteridae	0.10±0.37	-	-	-
		Psephenidae	-	-	0.03±0.06	0.02±0.04
		Ptilodactylidae	0.16±0.63	-	0.07±0.09	0.14±0.32
		Staphylinidae	0.02±0.04	-	0.39±1.04	0.08±0.12
	Collembola	Isotomidae	0.21±0.27	0.06±0.14	0.28±0.35	0.05±0.08
	Diptera	Ceratopogonidae	7.20±5.28	6.14±8.02	10.9±11.7	5.84±4.06
		Chaoboridae	0.07±0.15	-	0.07±0.21	-
		Chironomidae	77.43±50.15	50.57±44.11	105.6±41.8	74.56±41.89
		Corethrellidae	0.08±0.13	0.02±0.05	0.008±0.71	-
		Culicidae	0.18±0.22	0.03±0.08	0.43±0.02	0.24±0.61
		Dixidae	0.12±0.26	-	0.041±0.1	0.31±0.63
		Empididae	0.18±0.24	0.10±0.12	0.40±12.40	0.63±0.57
		Limoniidae	3.31±3.79	0.72±0.97	2.91±3.14	0.56±1.23
		Psychodidae	0.13±0.28	0.09±0.14	0.29±0.41	0.63±1.07
		Simuliidae	2.12±3.97	2.65±5.90	5.06±8.76	1.06±0.97
		Stratiomyidae	-	-	-	0.06±0.18
		Tabanidae	0.07±0.11	0.10±0.16	0.15±0.17	0.14±0.26
		Tanypodinae	12.73±9.81	9.14±6.69	21.61±9.87	15.63±12.43
		Tipulidae	0.60±1.77	0.16±0.25	0.31±0.32	0.66±1.25
	Ephemeroptera	Baetidae	8.03±8.27	2.48±3.43	13.31±13.55	4.11±3.23
		Caenidae	0.59±1.78	0.11±0.15	0.83±1.90	0.11±0.20

	Coryphoridae	-	-	0.06±0.13	0.03±0.03
	Ephemeridae	-	0.01±0.05	-	0.01±0.03
	Euthyplociidae	1.10±1.38	0.63±0.89	1.93±2.82	3.76±3.05
	Leptohyphidae	4.05±5.47	1.60±2.08	9.99±8.83	4.31±2.67
	Leptophlebiidae	9.55±7.96	3.80±3.65	22.44±14.80	11.82±10.11
	Polymitarcyidae	1.00±1.56	0.30±0.35	0.68±1.07	0.23±0.40
Heteroptera	Belostomatidae	-	-	-	0.02±0.03
	Corixidae	0.10±0.15	-	-	-
	Gelastocoridae	0.04±0.13	0.03±0.06	0.03±0.11	-
	Gerridae	0.15±0.28	0.02±0.06	0.07±0.16	0.01±0.03
	Helothrephidae	0.20±0.51	-	0.26±0.42	-
	Mesoveliidae	0.02±0.08	0.02±0.08	0.11±0.24	0.04±0.09
	Naucoridae	0.10±0.14	0.13±0.32	0.72±1.00	0.28±0.47
	Nepidae	0.03±0.07	-	0.02±0.04	-
	Notonectidae	0.03±0.08	-	0.03±0.06	-
	Veliidae	0.72±0.65	0.27±0.27	0.79±0.64	0.26±0.45
Lepidoptera	Pyralidae	0.32±0.32	-	1.14±1.45	1.66±3.14
Megaloptera	Corydalidae	0.04±0.06	0.04±0.07	0.58±0.97	0.03±0.06
	Sialidae	-	-	0.07±0.16	-
Odonata	Calopterygidae	-	-	0.63±1.52	0.27±0.45
	Coenagrionidae	0.24±0.30	0.07±0.10	0.05±0.09	0.14±0.15
	Corduliidae	3.33±1.84	0.76±0.47	2.87±5.64	0.67±0.37
	Gomphidae	0.69±0.67	0.37±0.47	0.77±0.84	0.33±0.22
	Heliocharitidae	0.15±0.21	-	0.08±0.09	-
	Lestidae	-	-	-	0.16±0.30
	Libellulidae	0.30±0.56	0.31±0.45	0.34±0.48	0.04±0.04
	Megapodagrionidae	0.42±0.47	0.38±0.73	1.29±1.94	0.72±0.70
	Perilestidae	0.18±0.44	0.10±0.27	0.08±0.12	-
	Plastytiscidae	-	-	0.10±0.32	0.75±1.22
	Polythoridae	-	-	0.40±1.26	0.24±0.32
	Protoneuridae	0.03±0.05	0.03±0.13	0.18±0.44	0.16±0.31
	Pseudostigmatidae	-	1.21±0.81	0.01±0.03	-
Plecoptera	Perlidae	0.97±1.14	-	1.36±0.94	2.48±1.66
Trichoptera	Calamoceratidae	0.13±0.27	0.13±0.40	0.15±0.24	0.21±0.52
	Ecnomidae	-	-	0.17±0.34	-
	Glossosomatidae	0.05±0.13	-	-	-
	Helicopsychidae	5.31±5.30	2.87±3.63	0.14±0.02	-
	Hydrobiosidae	-	-	-	0.03±0.09
	Hydropsychidae	4.44±3.90	2.83±2.73	14.66±12.95	6.65±4.93
	Hydroptilidae	1.01±1.46	1.23±2.72	2.83±5.03	3.98±4.24
	Leptoceridae	2.22±4.56	0.70±0.75	1.97±2.14	0.51±0.90
	Odontoceridae	0.77±1.27	0.50±1.60	0.23±0.28	0.18±0.27
	Philopotamidae	-	0.07±0.21	0.77±1.98	0.84±1.10
	Polycentropodidae	0.91±0.78	0.47±0.49	1.60±1.85	0.47±0.43

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Annexe V

RIVER RESEARCH AND APPLICATIONS

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A MULTIMETRIC MACROINVERTEBRATE INDEX FOR THE IMPLEMENTATION OF THE EUROPEAN WATER FRAMEWORK DIRECTIVE IN FRENCH GUIANA, EAST AMAZONIA

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ABSTRACT

Neotropical, overseas regions of Europe are subjected to the same water policy objectives as the continental ones but were overlooked during recent developments of bioindicators that fulfil the Water Framework Directive guidelines. We designed a macroinvertebrate-based multimetric index [*Indice Biotique Macroinvertébrés de Guyane* (IBMG)] to assess ecological health in remote headwater-small streams of French Guiana, Europe's only overseas region of continental South America. Invertebrates were sampled at 95 sites including reference and impacted river reaches, following a standardized protocol. Among the 102 biological metrics calculated from site-specific data, we selected metrics exhibiting the best trade-off between high discrimination efficiency, low specificity, low redundancy and high stability under reference conditions. The IBMG is composed of two taxonomic richness-based metrics, two abundance-based metrics, one trait-related metric and a diversity index (Shannon's entropy). Each metric was weighted by its discrimination efficiency. Using a test data set, we found that the IBMG was sensitive to the range of disturbances in French Guiana. Finally, comparing the IBMG with other indices developed in other neotropical countries reveals that, for several reasons, multimetric indices developed in the neotropics may perform well in the context of the data sets used to generate them but would certainly fail to be robust when used elsewhere. Copyright © 2015 John Wiley & Sons, Ltd.

KEY WORDS: anthropogenic perturbation; biological diversity; biomonitoring; neotropical rivers; reference conditions

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INTRODUCTION

Intended to reach a good ecological status for *all* surface waters by 2015, Europe's Water Framework Directive (WFD; European Council, 2000) has set up unified guidelines for the design and implementation of biological assessment tools in member states. After a decade of research intended to classify surface waters into biogeographic and geomorphological types, to agree upon reference (pristine) conditions for each type and to design biological indices that measure ecological health in terms of similarity to a reference state (Bailey *et al*., 1998), recent examples of WFD-compliant bioassessment tools in continental Europe can be found, for instance, in the works of Gabriels *et al*. (2010, Belgium), Kelly *et al*. (2012, Ireland) and Mondy *et al*. (2012, France). European overseas countries and territories (OCTs) and outermost regions (OMRs) occur in most biogeographic areas of the world, from polar to tropical ranges. Although inclusion in European Union (EU) policies may vary among OCTs and OMRs, most of these territories are subjected to WFD objectives. However, these regions were overlooked during the development of methods that fulfil WFD's requirements, at the point that most if not all of them still lack WFD-compliant tools. This is particularly true of the French overseas departments, for example, Martinique and Guadeloupe in the Caribbean (but see Touron-Poncet *et al*., 2014, for recent updates), Reunion Island in the Indian Ocean and French Guiana (FG) in the Eastern Amazon. Differences in climatic, biogeographic and geomorphological conditions, as well as differences in anthropogenic pressure, preclude the transposition of bioassessment tools developed in continental Europe to overseas regions. Clearly, biological traits (habitat preferences, sensitivity to pollution, etc.), species richness and numerical dominance do not compare among biogeographic regions. Finally, applied river research in many overseas regions suffers from a lack of taxonomic knowledge, so that ecologists are faced with minimal background on the distribution patterns of aquatic species. It is worth noting however that most OCTs and OMRs are small islands, so their freshwaters are expected to host depauperate faunas compared with1 their continental counterpart (Bass, 2003; Boulton *et al*., 2008). Nevertheless, the taxonomic issue is more pressing in species-rich continental areas of the tropics. For instance,

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very little is known about macroinvertebrate taxonomy and distribution in remote headwater streams of FG, East Amazonia. Importantly, biological monitoring methods are generally less expensive than chemical methods, and this could be important in South American countries.

In FG, like in other South American countries, small streams are located in forested areas and exhibit high ecological quality; some if not most of them have never been impacted by any human activity. Although FG's primary forest remains one of the least impacted of the world, gold mining and timber for wood products have strong impacts upon river ecosystems. A detailed description of the physical–chemical impacts of logging and gold mining in streams of FG was given by Dedieu *et al*. (2014). In particular, the annual gold output of FG is 60 times higher than 25 years ago (Hammond *et al*., 2007), and after the exploitation of large rivers, gold mining is now clearly shifting towards smaller inland streams (Hammond *et al*., 2007; Brosse *et al*., 2011). Unfortunately, no biological indices or any form of biological indicators are available yet for small streams of FG. Given recent economic development, there is, therefore, a pressing need to implement WFD-compliant biological assessment tools in FG, so that future water quality assessments and objectives can be established in light of current ecosystem health and economic plans. As part of the EU water policy, multimetric indices based on biological quality elements (BQEs; e.g. diatoms, benthic macroinvertebrates, fish and macrophytes) became the standard to evaluate ecosystem health. Multimetric indices assemble and weight different types of metrics (e.g. taxonomic richness, abundance and functional traits) that effectively respond to environmental heterogeneity and impairment and therefore provide more accurate assessment of ecosystem health than single metrics (Barbour *et al*., 1999; Hering *et al*., 2006, 2010). In an earlier study, Chandesris and Wasson (2005) have delineated hydro-ecoregions of FG based on geomorphological, hydrological and climatic data. In addition, Dedieu *et al*. (2015) have classified small streams of FG, based on extensive sampling of macroinvertebrate (the most widely used BQEs to date) and physical–chemical data. Importantly, these works allowed us to identify stream types and reference conditions for FG streams, thus providing background for the development of WFD-compliant biological indices.

In the present study, we take a step towards the implementation of the WFD in Europe's only overseas department of continental South America, by proposing a multimetric index based on river macroinvertebrate diversity. In order to address instructions for consistency in methods imposed by national environmental agencies, we mostly followed the methodology established in metropolitan France by Mondy *et al*. (2012), with adaptations inherent to biogeographic differences in community diversity, fundamental knowledge of species/population ecology and anthropogenic impacts. We assessed the efficiency of our new multimetric index on a test data set. Finally, we discuss our results in comparison with other multimetric indices recently developed in the neotropics with the broader aim to highlight gaps in knowledge about (bio)geographic differences in ecosystem structure and functioning, as well as human activities that influence them.

MATERIALS AND METHODS

Study area

This study was conducted in FG (surface area = 83 534 km^2), East Amazonia, from September 2011 to December 2012. The climate is tropical moist with 3000–3400 mm of yearly precipitation mainly distributed over 280 days. There is a major drop in rainfall (dry season) between September and December and another shorter and more irregular dry period in March. The maximum monthly temperature averages 33.5 °C (32.1–35.8 °C), and the monthly minimum averages 20.3 °C (19.7–21 °C). FG's stream systems are organized around seven large rivers (Maroni, Kourou, Mana, Sinnamary, Comté, Approuagues and Oyapock); however, 'small streams' (from headwaters to rivers with a depth of <1 m and a width of <10 m) represent ~80 000 km in total length, that is, 70–80% of all running waters in the region.

Field sampling

We sampled 95 sites, distributed over 76 small streams belonging to FG's main drainage basins (Figure 1). It should F1 be noted that the sampling effort was higher in the northern part of FG, owing to the difficulty to access southern FG. In this specific area covered by dense rainforest and without road networks, complex logistics limited our ability to sample a larger number of sites. We however managed to collect some samples from the main southern river basins. All sites were sampled during the dry season in 2011 and 2012 (September–December). Indeed, most remote sites were not accessible during the rainy season. In addition, human perturbation is detected less efficiently during high flows because of dilution effect. Additionally, we sampled 26 sites, including 14 sites subjected to human impacts (e.g. logging and gold mining). These new data sets were used as 'test' data sets.

Physical variables were chosen to describe the heterogeneity of the river bed substrate at each site. They were recorded directly in the field and accounted for the percentage composition of organic and mineral substrate types, using the standardized protocol by Souchon *et al*. (2000). These variables

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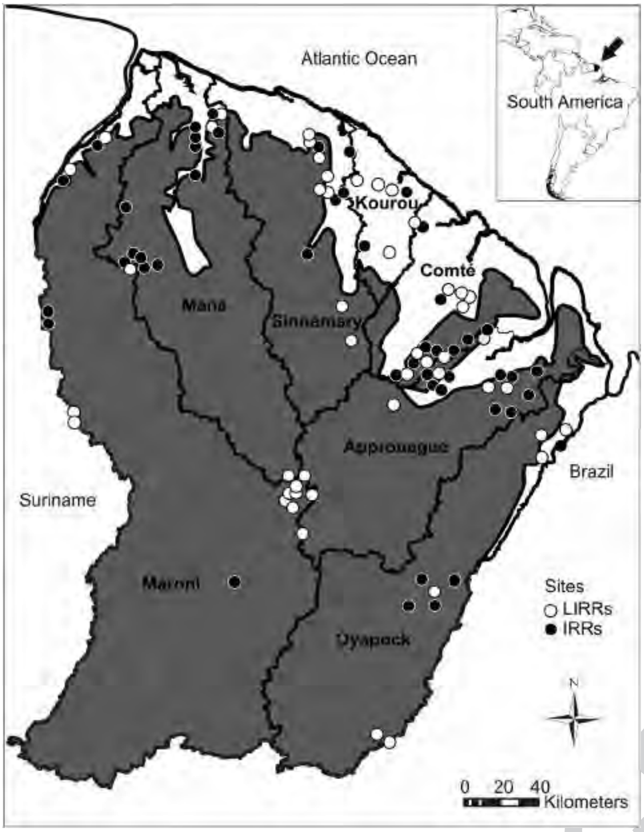

Figure 1. Location of the sampling sites in French Guiana. The two different shades illustrate the sub-regions (white: costal alluvial plain, grey: Guiana Shield) that form stream types according to hydro-ecoregions and macroinvertebrate assemblages (refer to text). Markers indicate the status of the sites: least impacted river reaches (LIRRs) and impacted river reaches (IRRs)

included percentage of leaf litter, percentage of submerged roots on the banks, percentage of submerged vegetation, mostly macrophytes, percentage of woody debris, percentage of silt, percentage of sand (<2 mm), percentage of gravel (2–25 mm) and percentage of coarse substratum (>25 mm). With coarse substrates being scarce in FG, this category of mineral substrate included pebbles, boulders and/or rocky outcrops.

Water samples for chemical analyses were taken at each site and immediately frozen. Chemical analyses were carried out at Hydreco Laboratory (Petit-Saut, FG) following standardized methods (AFNOR 2000, 2005a, 2005b). Chemical variables measured in the laboratory were total suspended matter (mg L^{-1}), nitrate (μg L^{-1}), total phosphorus (μg L^{-1}) and dissolved organic carbon (mg L^{-1}). Four variables were directly measured in the field using probes: dissolved oxygen (mg L^{-1}) (WTW 3205®), turbidity (NTU) (EUTECH®), pH (WTW 3110®) and conductivity (μS cm^{-1}) (WTW 3110®). Water temperature (°C) was the mean of values given by all aforementioned probes.

To collect the benthic macroinvertebrates, 12 sample units were taken at each site, that is, eight samples in organic substrates (group A samples, e.g. submerged vegetation and leaf litter) and four samples in mineral substrates (group B samples, e.g. sand and gravel). This distribution of samples was based on the aforementioned description of the various organic and mineral substrates in our small streams. Sample units in organic substrates consisted in intensive sweeping of a hand net (frame size = 46 × 23 cm; mesh size = 500 μm) during 1 min over a 0.46 × 1.5 m area (net width × 1.5 m). Sample units in mineral substrates were obtained by dragging a 5-cm layer of sediment with the same net, over a 0.46 × 1.5 m area. Prior to dragging, coarse substrata (pebbles) were brushed in front of the net and then removed. The samples were preserved in the field in 4% formalin (final concentration). Invertebrates were sorted in the laboratory and preserved in 70% ethanol. They were mostly identified to family (except for Annelida, Hydracarina, Nematoda and Planaria) and counted.

Stream types, least impaired river reaches and impaired river reaches

The ratios between observed biological parameters and the expected values under reference conditions (ecological quality ratios, discussed later) for these parameters are at the heart of WFD guidelines to evaluate river health (Hering *et al*., 2006). Hence, both typology and reference conditions need to be agreed upon before considering further developments. Chandesris and Wasson (2005) have established a typology of FG watersheds based on geomorphological, hydrological and climate data. A biological typology of streams based on benthic macroinvertebrates (Dedieu *et al*., 2015) confirmed Chandesris' conclusion that small streams can be clustered into two major sub-regions (i.e. into two stream types), namely the 'coastal alluvial plain' characterized by recent sediment and low elevations and the inland 'Guiana Shield' characterized by an eroded rocky substrate, a variability of elevations and large stream systems under a dense forest coverage.

Within each sub-region, we defined the status of each site [least impaired river reaches (LIRRs) versus impaired river reaches (IRRs)] based on National Survey Networks (DEAL Guyane, 2014), expert knowledge (e.g. presence of activities such as logging and gold mining was the most frequent criterion for IRRs), guidelines from the regional environmental agency and recent biological and physical–chemical data collected by us (Dedieu *et al*., 2014, 2015). Mann–Whitney tests have been applied to compare physical–chemical characteristics between LIRRs and IRRs.

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Metric set

We considered 102 metrics. These metrics can be divided into five categories: (1) taxonomic richness-related metrics (e.g. the number of species in a particular taxa group or a combination of taxa); (2) abundance-based metrics (e.g. the number of individuals); (3) diversity indices combining (1) and (2), for example, Shannon's entropy; (4) functional metrics (e.g. feeding habits); and (5) tolerance-related metrics (e.g. average score per taxon, Armitage *et al*., 1983). For each site, each metric was calculated on the basis of the following: (i) samples taken on organic substrates (A); (ii) samples taken on mineral substrates (B); and (iii) all samples (A + B) (Appendix I). Biological traits are poorly documented in stream invertebrates of FG compared with the European ones, so we selected five traits that are known at the family level for our fauna: functional feeding groups, locomotion, respiration, dispersal and habitat preferendum (Merritt and Cummins, 1996; Buss *et al*., 2004; Q3 Tomanova *et al*., 2006, 2008; Arrington and Winemiller, 2006; Ligeiro *et al*., 2010; Salman *et al*., 2013). Tolerance to pollution was based on the literature; for example, Elmidae and Ecnomidae were considered as sensitive taxa following Couceiro *et al*. (2007) and Lorion and Kennedy (2009). To quantify the sensitivity of taxa to water quality, the weighted average chemical pollution index of each taxon was calculated in order to determine the optimum value for a taxa (Ter Braak and Prentice 1988). These values ranged from 0 (low sensitivity to water quality degradation) to 5 (high sensitivity, Appendix II) and were used to calculate the average score per taxa.

Standardized effect size normalization, reference and worst values

In order to compare metric values obtained from different stream types, observed metric values were transformed into normalized deviations from values in reference conditions for a given stream type [standardized effect size (SES); Gotelli and McCabe, 2002]. SES normalization allowed us to identify the direction of deviation from values in LIRRs and allowed a direct comparison of metrics, regardless of river typology.

Standardized effect size values were calculated as follows:

$$SES = \left(Metric\_obs – mean_{ref}\right)/sd_{ref}$$

Metric_obs is the observed value of the metric, and $mean_{ref}$ and sd_{ref} are the average and standard deviation of the metric distribution under reference conditions for the same stream type.

Taking into account the discrimination efficiency (DE) of each variable (Ofenböck *et al*., 2004), we determined the type of response of each metric in impaired conditions. A given metric could exhibit three response patterns: (i) not responding significantly to the impairment (type 1), that is, the distribution of values from IRRs assemblages was not different from the distribution of values from LIRRs assemblages, and so neither $DE_{SES(25)}$ (proportion of IRR values smaller than the first quartile of the LIRR values distribution) nor $DE_{SES(75)}$ (proportion of IRR values higher than the third quartile of the LIRR values distribution) was higher than 0.25; (ii) decreasing with increasing impairment (type 2) [i.e. when $DE_{SES(25)} > 0.25$ and $DE_{SES(75)} < DE_{SES(25)}$]; and (iii) increasing with increasing impairment (type 3) [i.e. when $DE_{SES(75)} > 0.25$ and $DE_{SES(25)} < DE_{SES(75)}$] (Figure 2). Last, we determined F2 the reference values for each stream type and the worst value of each metric. The reference value corresponded to the highest (type 1 or 2) or lowest (type 3) value this metric could take in the LIRRs from a given stream type. The worst metric value corresponded to the lowest (type 1 or 2) or highest (type 3) value a metric could take in the IRRs from the whole data set. The 5th and the 95th percentiles of the distribution of values for a given metric were used as reference or worst values. This was performed in order to eliminate extreme metric values (Ofenböck *et al*., 2004).

Metric normalization

In order to identify similar patterns in metric responses to anthropogenic pressures for all stream types, and thus to facilitate the selection of metrics used at a large spatial scale, the observed values of the metrics were transformed into ecological quality ratios (EQRs) between observed and reference conditions for the same stream type at a time. EQRs were calculated following Hering *et al*. (2006), using formula 1 for metrics of types 1 and 2 and formula 2 for metrics of type 3:

$$EQR = (obs - lower)/(upper - lower) \tag{1}$$

$$EQR = 1 - (obs - lower)/(upper - lower) \tag{2}$$

with *obs* being the metric value observed for a given sampling point; *upper* and *lower* correspond to the *best* and *worst* value for this metric in the same stream type. In Equation 2, *upper* and *lower* correspond to the *worst* and *best* values of the metric. EQRs were bounded between 0 and 1. If observed values were greater than the best value (if quality is higher than the reference data set), the value of the EQRs was limited to 1. Conversely, if the values were smaller than the EQR worst value (if quality is lower than the worst values), the value of the EQR was fixed as 0.

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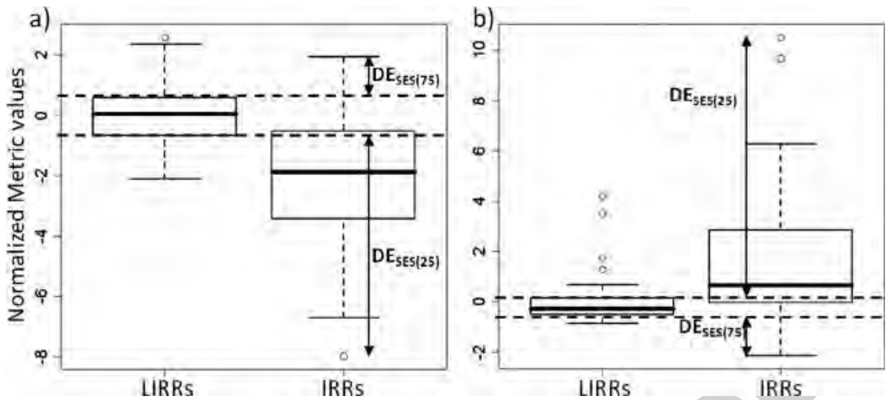

Figure 2. Boxplots of standardized effect size (SES) values of metrics in least impaired river reaches (LIRRs; left box) and impaired river reaches (IRRs; right box). (a) Discrimination efficiencies (DEs) of a type 2 metric (taxa richness); (b) DEs of a type 3 metric (GOLD index). Boxes delineate the 25th and 75th quartiles; thick lines represent the medians; circles are extreme values; whiskers extend to maxima and minima

Metric selection

Our aim was to determine metrics that best discriminate non-impacted sites from the impacted ones. We selected metrics that were expected to show the best trade-off between the following: (i) high DE; (ii) low specificity; and (iii) high stability under reference conditions (Mondy *et al*., 2012). The DE of a metric was calculated as the proportion of IRR assemblages with lower EQR values than the first quartile of the LIRR values distribution. The stability of a metric in reference conditions (i.e. LIRRs) was assessed using the coefficient of variation (CV) of EQR values distribution from LIRR assemblages. Robust estimates of DE and CV were obtained through a bootstrap procedure (mean of 100 calculations, each calculation using 60% of the sites randomly selected from the data set). We first selected the metrics that simultaneously exhibited a high DE and a high stability in LIRRs (average DE$\geq$0.5 and average CV in LIRRs$\leq$1/3).

Because we aimed at building a generalist index, metrics with a low specificity were preferred; that is, we selected metrics that were significantly correlated (linear regressions, $\alpha < 0.05$) to a high number of environmental variables reflecting site degradation. Thus, metrics significantly correlated to at least six water quality variables out of 17 were selected (see variables in Table I). To avoid redundancy, candidate metrics providing the same biological or ecological information (e.g. 'taxonomic richness in group A' and 'taxonomic richness in groups A + B') were gathered into homogenous groups. Only the metric with the lowest specificity and the highest DE for a given group of metrics was selected for possible inclusion in the multimetric index. Last, in order to limit the number of metrics, previously selected metrics were put into a correlation matrix. Metrics with >75% correlation were grouped by category (Section on Metric set), and we selected the best metrics (non-redundant in terms of bio-ecological information and higher DE) for each group.

Finally, the *Indice Biotique Macroinvertébrés de Guyane* (IBMG) was calculated using the following the equation:

$$IBMG = \frac{\sum DE_m \times EQR_m}{\sum DE_m}$$

with IBMG being the FG macroinvertebrate biotic index, DE_m the DE of the metric m and EQR_m the value of the metric m.

Ecological class boundaries and test of the Indice Biotique Macroinvertébrés de Guyane

In accordance with the WFD, we proposed five quality classes (i.e. 'high', 'good', 'moderate', 'poor' and 'bad' ecological quality). The identification of the ecological quality class boundaries was based on the distribution of the IBMG scores of the development data set. Most of the reference sites should be rated as good or very good in biological condition, as per WFD guidelines. The median and minimum values of the IBMG distribution in LIRRs were considered as the 'high–good' and 'good–moderate' boundaries, respectively (Figure 3). The 'bad–poor', 'poor–moderate' and 'good–high' boundaries were set using quartile and medians of the LIRR and IRR distributions (Figure 3). To evaluate the robustness of the IBMG DE, IBMG scores were calculated on the test data set (26 sites), and the DEs obtained with the development and test data sets were compared. Then, the stability of IBMG values in LIRRs was tested using two Kolmogorov–Smirnov tests. These tests were used to evaluate significant differences in the distribution of IBMG values from LIRRs between the test sets and the development data set.

All statistical procedures were performed with R software (R Development Core Team, 2009).

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Table I. Physical–chemical characteristics of the two stream types encountered in French Guiana

	Coastal alluvial plain					Guiana shield				
	LIRRs		IRRs			LIRRs		IRRs		
Variables	MN	SD	MN	SD	p	MN	SD	MN	SD	p
% Silt	8.04	9.04	48.24	21.43	**	8.61	6.82	34.00	20.63	**
% Sand	61.88	29.14	36.47	21.49	**	38.06	27.18	15.00	20.87	*
% Gravel	23.66	23.77	12.35	15.32	NS	19.12	15.45	19.17	14.78	NS
% Coarse substratum	6.88	10.98	2.94	5.32	NS	34.17	27.29	32.67	26.58	NS
% Woody debris	16.34	14.34	14.71	11.52	NS	20.14	19.45	7.33	6.08	*
% Submerged vegetation	10.45	11.39	20.15	21.02	*	1.67	4.11	1.67	3.62	NS
% Leaf litter	24.02	21.20	25.29	14.06	NS	17.89	13.89	13.83	17.85	*
% Submerged roots on the banks	20.26	16.51	9.26	7.33	NS	24.61	23.06	7.83	8.81	*
pH	5.29	0.54	5.33	0.31	*	6.19	0.79	6.38	0.38	NS
Water temperature (°C)	24.76	1.21	25.49	1.22	NS	24.36	1.63	25.64	1.91	*
Conductivity ($\mu S\,cm^{-1}$)	22.25	5.05	26.47	15.24	NS	39.88	29.28	48.93	31.99	NS
Dissolved oxygen ($mg\,L^{-1}$)	6.02	0.93	5.71	0.85	NS	6.96	0.58	6.84	0.59	NS
Turbidity (NTU)	2.06	1.24	6.52	8.60	*	3.16	2.23	54.70	92.20	**
Nitrate ($\mu g\,L^{-1}$)	0.30	0.15	0.24	0.12	NS	0.23	0.14	0.30	0.13	NS
Total phosphorus ($\mu g\,L^{-1}$)	0.02	0.02	0.06	0.04	*	0.04	0.03	0.02	0.02	NS
Total suspended matter ($mg\,L^{-1}$)	6.32	6.09	12.03	14.68	*	3.19	2.05	58.79	143.38	*
Dissolved organic carbon ($mg\,L^{-1}$)	22.04	16.69	20.31	11.64	NS	13.23	9.87	12.27	6.23	NS

IRRs, impacted river reaches; LIRRs, least impacted river reaches; MN, mean; SD, standard deviation; NS, no significant difference.
*$p < 0.05$ and **$p < 0.01$ (Mann–Whitney tests).

RESULTS

Environmental variables

Regardless of stream types, some environmental variables differed significantly between LIRRs and IRRs. Compared with the reference sites, impaired sites had significantly higher values for turbidity, amount of total suspended matter and percentage of silt (Table I). In the coastal alluvial plain, reference sites had lower total phosphorus concentration and percentage of submerged vegetation than the impacted ones. Within the Guiana Shield area, impacted streams accumulated lower amounts of woody debris and leaf litter and had lower percentage of submerged roots on the banks, compared with the reference sites. Water temperature was also slightly higher at these impacted sites, because of clearing of the riparian forest (Dedieu *et al*., 2014).

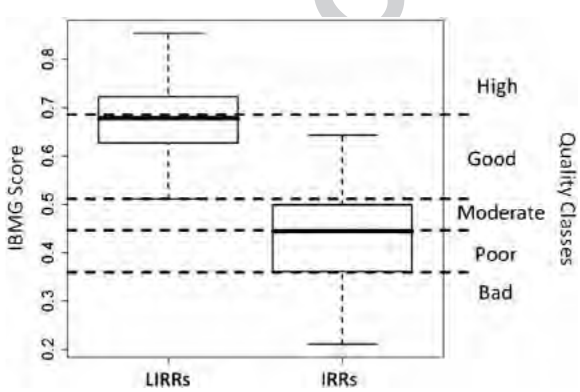

Figure 3. Ecological quality class boundaries (black dashed lines) for the *Indice Biotique Macroinvertébrés de Guyane* (IBMG). Boxplots represent the IBMG score distributions in least impaired river reaches (LIRRs; left box) and impaired river reaches (IRRs; right box) ranging from the 25th to 75th percentiles of the distribution; thick lines represent the medians; circles are extreme values; whiskers extend to maxima and minima

Metric selection and index construction

Among the 102 metrics tested on the different sample groups (A, B and A + B), 56 differed significantly between LIRRs and IRRs in at least one sample group (Appendix A). Then, 36 metrics exhibited a mean DE greater than 0.5 and an average CV in LIRRs ≤ 1/3 (Table II). Seventeen of these 36 metrics significantly responded to at least six environmental variables (out of 17). From these 17 metrics, we eliminated redundant information by selecting the most discriminant metrics among the ones that provided similar bio-ecological information. After considering pairwise correlations of metrics, 12 metrics were eliminated. The Shannon index exhibited a DE slightly inferior to 0.5 (DE = 0.47 and CV = 0.10); it was however added to the final multimetric index to fulfil recommendations of our national environmental agencies. Hence, the IBMG was finally composed of six metrics: Chao1 (B), Log.Elmidae (A), Number of Coleopteran families (A + B), %collector-gatherer (A + B), %Ephemeroptera

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Table II. Mean DE, mean CV and responses of 36 candidate metrics to the 17 environmental variables listed in Table I

Candidate metrics	Description	DE	CV	*N*
Margalef_B	Margalef index calculated on group B samples	0.79	0.19	3
Chao1_AB	Chao estimator 1 calculated on groups A and B samples	0.76	0.11	10
TaxaS_A	Taxa richness calculated on group A samples	0.76	0.12	5
TaxaS_AB	Taxa richness calculated on groups A and B samples	0.73	0.11	10
Chao1_A	Chao estimator 1 calculated on group A samples	0.72	0.12	5
Chao1_B	Chao estimator 1 calculated on group B samples	0.69	0.28	7
TaxaS_B	Taxa richness calculated on group B samples	0.69	0.26	5
Log.Elmidae_A	Logarithm of the abundance of Elmidae calculated on group A samples	0.69	0.32	7
Margalef_AB	Margalef index calculated on groups A and B samples	0.69	0.18	10
Margalef_A	Margalef index calculated on group A samples	0.65	0.17	2
Brillouin_A	Brillouin index calculated on group A samples	0.63	0.13	7
SwimQ_AB	% Swimmers calculated on groups A and B samples	0.63	0.32	5
CoGaQ_A	% Collector-gatherers calculated on group A samples	0.61	0.17	6
ETQ_AB	% Ephemeroptera and Trichoptera calculated on groups A and B samples	0.60	0.25	2
Simpson_B	Simpson index calculated on group B samples	0.60	0.28	2
EPTLQ_A	% Ephemeroptera, Plecoptera, Trichoptera and Lepidoptera calculated on group A samples	0.60	0.27	7
EPTQ_A	% Ephemeroptera, Plecoptera and Trichoptera calculated on group A samples	0.59	0.30	3
EPTLMQ_AB	% Ephemeroptera, Plecoptera, Trichoptera, Lepidoptera and Megaloptera calculated on groups A and B samples	0.59	0.24	6
ColeoS_AB	Coleoptera family richness calculated on groups A and B samples	0.59	0.25	6
TrichoS_AB	Trichoptera family richness calculated on groups A and B samples	0.59	0.17	4
ColeoS_A	Coleoptera family richness calculated on group A samples	0.59	0.26	1
CoGaQ_AB	% Collector-gatherers calculated on groups A and B samples	0.59	0.17	9
ETQ_A	% Ephemeroptera and Trichoptera calculated on group A samples	0.59	0.30	2
EPTQ_AB	% Ephemeroptera, Plecoptera and Trichoptera calculated on group B samples	0.58	0.24	1
EPTS_B	Ephemeroptera, Plecoptera and Trichoptera family richness calculated on group B samples	0.56	0.12	0
Falpha_A	Fisher's alpha index calculated on group A samples	0.56	0.30	2
Brillouin_AB	Brillouin index calculated on groups A and B samples	0.54	0.16	5
PMS_A	Plecoptera and Megaloptera family richness calculated on group A samples	0.54	0.21	3
ClingQ_AB	% Clingers calculated on groups A and B samples	0.54	0.32	3
ShannonH_A	Shannon index calculated on group A samples	0.53	0.17	9
EPTLMS_B	Ephemeroptera, Plecoptera, Trichoptera, Lepidoptera and Megaloptera family richness calculated on group B samples	0.53	0.11	1
Simpson_A	Simpson index calculated on group A samples	0.53	0.07	4
TrichoS_B	Trichoptera family richness calculated on group B samples	0.53	0.20	0
ETS_B	Ephemeroptera and Trichoptera family richness calculated on group B samples	0.52	0.12	1
TrichoS_A	Trichoptera family richness calculated on group A samples	0.51	0.19	1
ShannonH_AB	Shannon index calculated on groups A and B samples	0.47	0.10	5

DE, discrimination efficiency; CV, coefficient of variation; *N*, number of environmental variables significantly correlated to the metrics.

and Trichoptera (A + B) and the Shannon index (A + B). The DE and CV in LIRRs of the IBMG were 0.79 and 0.15, respectively. The best value of each stream type and the worst values of the data set needed to calculate the EQR of each metric are given in Appendix C.

Ecological quality class boundaries and index test

The values of the IBMG scores of the development data set ranged from 0.21 to 0.85 and were used to set the quality class boundaries (Figure 3). The 'good' lower boundary was set at 0.51 so that all reference sites were included in it (Barbour *et al*., 1999). The high–good and moderate–poor boundaries corresponding to the medians of the LIRR and IRR distributions and were set at 0.69 and 0.45, respectively. The poor–bad boundary was set at 0.36 corresponding to the 25th quartile of the distribution of IRR scores. Values for the DE and CV in LIRRs calculated with the test data set were 0.83 and 0.11, respectively. The distributions of IBMG scores in LIRRs of the development and test data sets showed no significant difference (Kolmogorov–Smirnov test: $D = 0.1913$, $p = 0.8708$).

DISCUSSION

In this study, we propose a multimetric index for the biological assessment of East Amazonian streams, developed under an EU framework. We assembled metrics that respond to several environmental variables associated with

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ecosystem impairment. Therefore, the IBMG can be considered as a generalist index in that it responds to the main types of pressures encountered in FG. Some studies showed that there is no significant difference in assemblage structure between dry and rainy seasons (Baptista *et al*., 2007; Couceiro *et al*., 2012). For practical reasons, the IBMG is based on samples taken during the dry season. Nevertheless, most remote sites cannot be reached (and therefore monitored) during the rainy season, and environmental managers will chiefly implement bioassessment campaigns during the dry season only. The IBMG can also be viewed as a preliminary tool in that it is based on a single campaign, and the test data set is limited to 26 sites. However, future surveys are expected to provide further data on the same network of sites during the next years, and this will allow testing and refining of the index. (Figure 4)

The six selected metrics account for effects of anthropogenic impairment on different environmental factors. Log. Elmidae and %collector-gatherers responded to changes in percentage of woody debris, percentage of submerged vegetation and percentage of submerged roots, a series of organic habitat variables that indirectly account for the riparian habitat quality (Compin and Céréghino, 2007). Chao1, Shannon index, the percentage of Trichoptera–Ephemeroptera and the number of coleopteran families mostly correlated with mineral particle size (percentages of sand, gravel and coarse substratum) and water chemistry (turbidity, total suspended matter and total phosphorus). These metrics therefore did well at accounting for changes in the instream physical–chemical habitat quality. The fact that both A and B sample groups are included in the calculation of most metrics evidences that the structure of macroinvertebrate assemblages varied at the reach scale and that this variability is relevant for identifying the impaired status of streams in FG (Buss *et al*., 2004). Two metrics out of six are calculated on a group of samples only (A or B), thus reducing variability

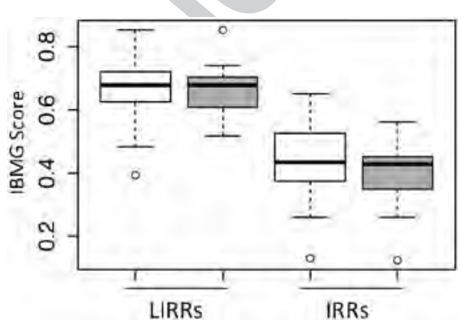

Figure 4. Distribution of the *Indice Biotique Macroinvertébrés de Guyane* (IBMG) scores in least impaired reaches (LIRRs) and impaired river reaches (IRRs) for the development data set (white boxes) and the test data sets (grey boxes). Rectangles delineate the first and third quartiles; the thick lines represent the medians; circles are outliers; bars are maxima and minima

in the reference condition samples (LIRRs). Although the Chao1 estimator on sample group A + B had a higher DE and exhibited higher stability in reference conditions, it finally appeared to be highly correlated with the other candidate metrics. We thus selected the Chao1 estimator on sample group B instead. Our index performed well in separating reference and disturbed sites because 79% of the sites that were *a priori* classified as impaired were assigned to categories below 'good' quality by the IBMG. Testing the index with an independent data set also allowed us to demonstrate its stability in LIRRs and its robustness in terms of DE. Still, future works could improve the IBMG. Specifically, metrics able to detect diffuse pollution from the sparse human settlements or small cultivations are probably lacking, but these particular pressures are scarce in FG and could not be found with a sufficient replication in this study.

In other neotropical countries, multimetric indices were recently developed under national frameworks (summarized in Table III). Interestingly, we note differences in selected metrics among countries. These differences might therefore be due to the following: (i) biogeographic differences in community structure; (ii) differences in anthropogenic pressure types and/or intensity; and/or (iii) differences in methods to design a multimetric index. Assuming that the reference condition approach (Bailey *et al*., 1998) has become a worldwide standard to design biotic indices, one may expect that biogeographic differences in species composition and traits, and to some extent differences in anthropogenic pressures, primarily account for differences in metric assemblages across countries. Taxonomic resolution is also an important issue in biological assessments (Waite *et al*., 2004), and most if not all rapid bioassessment methods rely on family or family–genus identifications of benthic macroinvertebrates (Heino, 2014). In recent neotropical studies, only Touron-Poncet *et al*. (2014) used species-level identifications of aquatic insects in Caribbean islands—but in this insular context, most genera only had one species. On the methodological side, however, multimetric indices in the neotropics were designed for study areas ranging from a few hundred (Couceiro *et al*. 2012) to about one million square kilometres (Moya *et al*., 2011; Villamarín *et al*., 2013) and for stream types ranging from first to second orders (this study) to first to fifth orders (Oliveira *et al*., 2011) (Table III). Differences in spatial and longitudinal scales have a significant influence on the natural variability of communities, thereby fostering the selection of certain metric types. For instance, whereas particular taxa are more likely to respond to disturbance at small geographic scales (e.g. Baptista *et al*., 2007; Touron-Poncet *et al*., 2014), a larger number of metrics that are independent of taxonomy (e.g. functional groups and biological traits) should theoretically be selected at larger scales (Moya *et al*., 2011). Biogeography-related differences in final metric assemblage

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Table III. Comparison of multimetric indices recently developed in the neotropics under the reference condition approach

Reference	Region; stream types	Study area (km^2)	Main disturbance type(s)	Metrics selected	ID level
This study	French Guiana; first-order to second-order streams	83 534	Gold mining; deforestation	Chao1 estimator Shannon index Coleoptera richness Log(Elmidae abundance) % Collector-gatherers % ET Preference for silt	Family
Touron-Poncet *et al*. (2014)	Martinique and Guadeloupe islands; first-order to third-order streams	1718	Urban, domestic and industrial runoff; WWTP; agriculture	Preference for boulders Abundance of Ephemeroptera ETC richness Trichoptera Richness Shannon index Taxa richness	Genus and species
Villamarín *et al*. (2013)	Ecuador and Peru; first-order to third-order streams	1 568 736	Agriculture, livestock grazing; urban sewage	Richness of intolerant taxa EPT richness % clingers % climbers % tolerant taxa Taxa richness	Genus
Helson and Williams (2013)	Panama; third-order to fourth-order streams	1050	Agriculture, urban sewage	Margalef index Shannon index % EPT % Trichoptera Chironomidae : Diptera ratio % Scrapers % Shredders	Genus
Couceiro *et al*. (2012)	Brazil, central Amazon; first-order to second-order streams	<1000	Deforestation, urban sewage	Taxa richness (family) EPT richness % EPT EPT : Chironomidae ratio Richness of sensitive taxa % Collector-gatherers % Shredders	Genus
Moya *et al*. (2011)	Bolivia; first-order to fourth-order streams	1 098 581	Urban sewage, agriculture, mining, deforestation	Taxa richness (family) EPT richness % deposit feeders % microphyte feeders % coarse detritus feeders % flat-bodied taxa Total abundance % EPT	Genus
Baptista *et al*. (2007)	Brazil, Southeastern Atlantic forest; first-order to third-order streams	12 904	Urban sewage, agriculture	% Diptera % Coleoptera Taxa richness (family) EPT Richness BMWP index (adapted) % Shredders	Genus
Oliveira *et al*. (2011)	Brazil, Rio de Janeiro State; first-order to fifth-order streams	1265	Urban sewage	Taxa richness (family) Trichoptera richness Shannon index % Plecoptera % EPT % Mollusca + Diptera % Shredders Hydropsychidae : Trichoptera ratio Chironomidae: Diptera ratio (%)	Genus

(*Continues*)

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Table III. (Continued)

Reference	Region; stream types	Study area (km^2)	Main disturbance type(s)	Metrics selected	ID level
Ferreira *et al.* (2011)	Southeastern Brazil; third-order to fourth-order streams	29 173	Industry, mining, urban sewage, damming	Taxa richness (family) % Oligochaeta % Chironomidae + Oligochaeta % EPT % Collector-gatherers BMWP index (adapted)	Family
Suriano *et al.* (2011)	Brazil, Sao Paulo State; first to second orders	248 800	Urban sewage, agriculture	Family richness EPT richness % EPT % Megaloptera + Hirudinea Shannon index (genus level) BMWP index (adapted)	Genus

WWTP, wastewater treatment plants; BMWP, biological monitoring working party; ID, identification.

can also be detected. For instance, Plecoptera richness or relative abundance commonly form metrics either *per se* or in combination with other sensitive taxa (Trichoptera and Ephemeroptera). This indicator group is not relevant in most of the neotropics where only one genus (*Anacroneuria*) is known (Fenoglio and Rosciszewska, 2003), and in Caribbean islands where it is entirely absent. These examples show that within a vast biogeographical area such as the neotropics, biological indicators developed for a given sub-area or country cannot be easily transposed to other geographic areas. Considering the influence of pressure types on assemblages of metrics, taxa that indicate water pollution (Chironomidae, Oligochaeta and Hirudinea) were more commonly selected in areas subjected to urban and agricultural pollution (e.g. Ferreira *et al*., 2011; Suriano *et al*., 2011), two types of disturbance that are infrequent in FG. Conversely, taxa with very specific requirements as regards mineral particle size do well at revealing degradation of the physical habitat (gold mining and deforestation in FG). This is particularly true of Trichoptera, especially in case-building taxa that require sand and piece of woods to build their larval case. Such selected examples support the hypothesis that freshwater neotropical communities depend not only on water quality but also on habitat components.

The IBMG fulfils the WFD requirements of taking into account the abundance and diversity of taxa. The inclusion of biological traits is highly desirable (Mondy *et al*., 2012), but unfortunately, biological information is lacking in tropical areas where the autoecology of most species is poorly (or not) documented (Tomanova, 2007; Moya *et al*., 2011). Still, the IBMG includes a trait-related metric. Another requirement is that ecological evaluation should regard sensitivity of taxa to pollution (European Council, 2000). To date, tolerance values are lacking for the regional fauna, and several studies have cautioned that tolerance values in the temperate regions do not apply to the neotropics (Moya *et al*., 2007). Initially, metrics indicating tolerance of taxa were calculated (Materials and Methods section), but they were not selected by our statistical procedures. Nevertheless, Ephemeroptera, Trichoptera and to a lesser extent Coleoptera are considered as sensitive to pollution by stream ecologists. Thus, the use of these taxa in our metrics at least partially take into account the 'pollution sensitivity' of taxa within assemblages. Therefore, we can reasonably consider that the index matches this last WFD criterion. Finally, comparing the IBMG with other indices reveals that, for several non-mutually exclusive reasons, multimetric indices developed in the neotropics may perform well in the context of the data sets used to generate them but would certainly fail to be robust when used elsewhere. Ideally, there would thus be a need to intercalibrate these indices in an attempt to harmonize operational practices and reach a biogeographic region-wide biological assessment scheme.

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APPENDIX A

The 102 metrics tested to build the multimetric index and the expected and observed responses to the stressor gradient. The expected responses were based on information reported in the literature (Merritt and Cummins, 1996; Moya *et al*., 2011; Helson and Williams, 2013; Villamarín *et al*., 2013). Mann–Whitney test compare metric values among LIRRs and IRRs: *$p < 0.05$; **$p < 0.01$; ***$p < 0.005$; ns, not significant.

Candidate metrics	Expected response	Observed response	*p*-values between samples		
			A	B	A + B
Abundance					
Total number of individuals	Decrease	Decrease	*	*	**
% Coleoptera	Decrease	Decrease	***	ns	***
% Crustacea	Decrease	Decrease	ns	ns	ns
% Diptera	Increase	Increase	ns	ns	ns
% Ephemeroptera	Decrease	Decrease	**	ns	**
% Hemiptera	Variable	Decrease	*	ns	*
% Odonata	Variable	Decrease	ns	**	ns
% Oligochaeta	Increase	Increase	**	*	**
% Trichoptera	Decrease	Decrease	ns	*	*
% Planaria	Variable	Decrease	ns	ns	ns
% Lepidoptera	Decrease	Decrease	ns	ns	ns
% EPT	Decrease	Decrease	**	*	**
% ET	Decrease	Decrease	***	**	***
% EPT/Diptera	Decrease	Decrease	ns	ns	ns
% ET/Diptera	Decrease	Decrease	ns	ns	ns

(Continues)

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Table . (Continued)

Candidate metrics	Expected response	Observed response	p-values between samples		
			A	B	A + B
% ET/Diptera + Oligochaeta	Decrease	Decrease	ns	ns	ns
% EPTLM	Decrease	Decrease	ns	***	ns
% Gasteropoda + Oligochaeta + Diptera (GOLD)	Increase	Increase	*	ns	*
% Non-insects	Increase	Increase	ns	ns	ns
% Hydropsychidae/Trichoptera	Decrease	Decrease	ns	ns	ns
% Polycentropodidae	Increase	Decrease	ns	ns	ns
% Polycentropodidae/Trichoptera	Increase	Decrease	ns	ns	ns
% Leptophlebiidae/Leptohyphidae	Decrease	Variable	ns	*	ns
% Baetidae/Ephemeroptera	Increase	Increase	ns	ns	ns
% Caenidae/Ephemeroptera	Increase	Variable	*	ns	ns
% Ceratopogonidae	Increase	Increase	*	ns	*
% Tanypodinae/Chironomidae	Increase	Increase	*	ns	**
% Ceratopogonidae + Tanypodinae/Chironomidae	Increase	Increase	ns	ns	ns
% Chaoboridae, Corethrellidae, Culicidae	Decrease	Decrease	*	ns	*
% Notonectidae	Increase	Increase	ns	ns	ns
% Notonectidae/Hemiptera	Increase	Increase	ns	ns	ns
Diversity and evenness indices					
Dominance *D*	Increase	Increase	*	ns	ns
Simpson	Variable	Decrease	*	ns	ns
Shannon *H*	Decrease	Decrease	**	*	*
Evenness $e^{H/S}$	Decrease	Decrease	**	*	*
Brillouin	Decrease	Decrease	***	***	***
Menhinick	Decrease	Decrease	**	*	*
Margalef	Decrease	Decrease	***	***	***
Equitability *J*	Decrease	Decrease	ns	ns	ns
Fisher alpha	Decrease	Decrease	***	*	**
Berger–Parker	Decrease	Decrease	**	*	*
Chao-1	Decrease	Decrease	**	***	**
Taxonomic richness					
Taxa richness	Decrease	Decrease	***	***	***
Family richness	Decrease	Decrease	***	***	***
Number of Coleoptera families	Decrease	Decrease	***	**	***
Number of Crustacea families	Decrease	Decrease	ns	ns	ns
Number of Diptera families	Increase	Variable	*	ns	ns
Number of Ephemeroptera families	Decrease	Decrease	***	**	***
Number of Hemiptera families	Decrease	Variable	ns	ns	ns
Number of Odonata families	Decrease	Decrease	ns	*	ns
Number of Trichoptera families	Decrease	Decrease	**	**	**
Number of EPT families	Decrease	Decrease	**	ns	*
Number of EPT/Diptera families	Decrease	Decrease	ns	ns	ns
Number of ET families	Decrease	Decrease	*	*	*
Number of EPTLM families	Decrease	Decrease	*	*	ns
Sensitivity to water pollution					
% Stratiomyidae + Oligochaeta	Increase	Increase	*	ns	**
ASPTg (tolerance values in French Guiana)	Decrease	Decrease	**	**	*
% Euthyplociidae	Decrease	Decrease	**	ns	ns
% Tabanidae	Decrease	Decrease	*	ns	ns
% Megaloptera	Decrease	Increase	ns	ns	ns
% Polythoridae	Decrease	Decrease	ns	ns	ns
% Dixidae	Decrease	Decrease	*	ns	ns
% Perilestidae	Decrease	Decrease	*	*	*
% Perlidae	Decrease	Decrease	ns	ns	ns
% Leptoceridae	Decrease	Decrease	ns	*	ns
% Elmidae	Decrease	Decrease	***	ns	**
% Elmidae/(Coleoptera)	Decrease	Decrease	ns	ns	ns
Log.Elmidae	Decrease	Decrease	***	*	***

(Continues)

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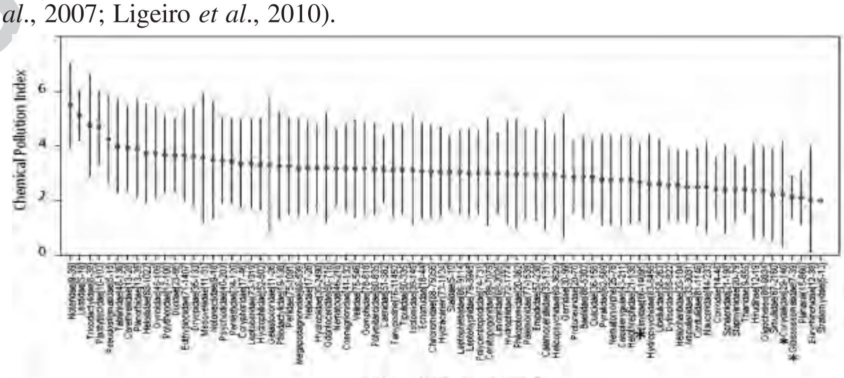
Table . (Continued)

Candidate metrics	Expected response	Observed response	*p*-values between samples		
			A	B	A + B
% Ecnomidae	Decrease	Decrease	ns	**	ns
% Plastytiscidae	Decrease	Decrease	ns	*	ns
% Lestidae	Decrease	Decrease	ns	ns	ns
Functional groups					
% Predators	Decrease	Variable	ns	ns	ns
% Scrapers	Variable	Decrease	ns	*	ns
% Collector-filterers	Decrease	Variable	*	*	*
% Collector-gatherers	Variable	Decrease	*	**	*
% Parasites	Increase	Increase	ns	ns	ns
% Predators/non-predators	Decrease	Variable	ns	ns	*
Number of predator families	Decrease	Decrease	ns	ns	ns
Number of collector-gatherer families	Increase	Decrease	*	**	*
Number of collector-filterer families	Decrease	Decrease	ns	ns	ns
% Surface swimmers	Decrease	Decrease	*	*	*
% Open-water swimmers	Variable	Variable	*	*	*
% Clingers	Variable	Decrease	ns	*	ns
% Burrowers	Increase	Increase	ns	ns	ns
% Climbers	Decrease	Decrease	ns	ns	ns
% individuals using respiration with tegument	Increase	Increase	ns	*	*
Number of taxa using respiration with tegument	Increase	Increase	ns	ns	ns
% Individuals with gills	Decrease	Decrease	ns	*	ns
Number of taxa with gills	Decrease	Decrease	ns	ns	ns
% Individuals using respiration with plastron	Variable	Decrease	ns	*	ns
Number of taxa using respiration with plastron	Variable	Decrease	ns	ns	ns
% Piercer	Increase	Variable	ns	ns	ns
% Detritivorous	Variable	Decrease	ns	ns	ns
% Individuals with protected gills	Increase	Increase	ns	ns	ns
% Taxa with preference for sand	Increase	Increase	ns	**	ns
% Taxa with preference for bank	Variable	Decrease	*	ns	ns
% Taxa with preference for stones	Decrease	Decrease	ns	ns	ns
% Taxa with preference for litter	Variable	Increase	*	ns	ns
% Flight dispersers	Decrease	Decrease	ns	ns	ns
Number of taxa using flight dispersal	Decrease	Decrease	ns	ns	ns
% Individuals using passive dispersal	Variable	Increase	ns	ns	ns
Number of taxa using passive dispersal	Variable	Increase	ns	ns	ns

APPENDIX B

Chemical pollution index (CPI) values of taxa. Mean (squares) and standard deviation (lines) of CPI are weighted by the abundance of taxa. Taxa presented were identified to family and were present in at least seven samples with over 50 specimens. An asterisk indicates three families with low values obtained by the CPI but were considered as sensitive to pollution based on the literature (Couceiro *et al*., 2007; Ligeiro *et al*., 2010).



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APPENDIX C

Reference and worst values used for the normalization of the six IBMG individual metrics according to stream types.

Reference values	Chao1_B	Shannon_AB	CoGa.Q_AB	Coleo.S_AB	Log.Elmidae_A	ET.Q_AB
Costal alluvial plain						
Upper	1.945	1.256	1.189	1.485	1.735	1.362
$mean_{ref}$	22.533	2.323	63.955	12.755	0.995	21.091
sd_{ref}	5.890	0.405	8.864	3.231	0.329	8.0766
Guiana Shield						
Upper	1.547	1.398	1.518	1.296	1.545	1.749
$mean_{ref}$	28.50	2.346	63.780	11.771	0.945	22.146
sd_{ref}	7.018	0.305	10.123	3.536	0.353	5.374
Lower	−3.395	−4.189	−4.130	−2.773	−2.786	−3.682

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Annexe VI

Ecological Indicators 52 (2015) 332–340

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Assessing the impact of gold mining in headwater streams of Eastern Amazonia using Ephemeroptera assemblages and biological traits

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ABSTRACT

Whilst the biological traits composition of invertebrate assemblages has been successfully used to monitor temperate rivers, it has been seldom tested in tropical areas. We compared the trait composition of Ephemeroptera assemblages (five traits, 21 modalities) in three categories of headwater streams of FG: reference (undisturbed) sites, sites formerly impacted by gold-mining, and sites currently impacted by gold-mining. Differences in macroinvertebrate assemblage according to environmental characteristics and disturbance were evaluated using correspondence analysis and MANOVA. Among the considered traits, food acquisition, respiration and locomotion detected both past and current disturbance associated with gold-mining in headwaters. A fuzzy correspondence analysis showed a significant segregation of currently gold-mined, formerly gold-mined, and reference sites according to species traits. Shifts in trait composition were mostly related to changes in assemblage composition. Interestingly, no significant decline in diversity indices was observed in formerly gold-mined sites compared to the reference sites, 2 years after abandonment, while the taxonomic and trait composition of communities changed at these sites. These results support the case for further fundamental quantification of species traits, and for the inclusion of sensitive, trait-related metrics in upcoming multimetric indices for the assessment of river health.

1. Introduction

Throughout the world, environmental legislation aiming at surveying, managing and protecting freshwater ecosystems relies on biological indicators of ecosystem health (Stoddard et al., 2008; Dos Santos et al., 2011). For instance, regional-national surveys of stream systems provide large volumes of site-specific data on biological communities and the associated physical–chemical environments (e.g., Harris and Silveira, 1999a,b; Paulsen et al., 2008; Mondy et al., 2012). The ecological health of rivers is then defined in terms of deviation from a *reference* state where human impacts are almost null (Bailey et al., 1998). Biological traits of freshwater organisms (e.g., body size, feeding habits, etc.) are potentially more useful than taxonomic structure (species × abundance data) to detect patterns of deviation from reference conditions where different hydroecoregions (areas that differ by geology, climate, vegetation, and species composition) are covered (Bonada et al., 2006), though ecologists traditionally make use of taxa lists (e.g., Bernadet et al., 2013). Whilst species occurrence may have a strong stochastic element and local-regional validity only, traits reflect environmental conditions and may be shared among many species (Southwood, 1988; Statzner et al., 2001). Traits may give greater insight into habitat change (Dolédec et al., 1999; Statzner et al., 2004) and their determination generally requires less taxonomic expertise (Dolédec et al., 2000), so that it can be utilized where limited information is available, and/or for animal groups where taxonomic knowledge is limited.

Most applications of biological traits to bioassessment were developed in the temperate zone, and were based on benthic macroinvertebrates (e.g., Usseglio-Polatera et al., 2000; Gayraud et al., 2003; Statzner et al., 2005; Dolédec et al., 2006). This is owing to the fact that species traits are poorly documented in tropical invertebrates compared, for instance, to the European or North-American ones (Touron-Poncet et al., 2014). To the best of our knowledge, only a few studies used a biological traits approach in tropical rivers (Tomanova, 2007; Tomanova et al., 2008) to assess how macroinvertebrate community functions change along gradients of anthropogenic disturbance. Recent efforts on the taxonomy

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of the South American Ephemeroptera (mayflies) have provided valuable information on the diversity of this insect order in the neotropics (Heckman, 2002; Salles et al., 2004; Domínguez et al., 2006; Chacón et al., 2009), and in addition, Ephemeroptera are recognized as relevant biological indicators, because of their sensitivity to a wide array of disturbance types (Landa and Soldan, 1991; Buffagni, 1997). Their taxonomic richness and/or abundance have notably proven relevant parameters for the design of multimetric indices in temperate (Gabriels et al., 2010) and neotropical (Couceiro et al., 2012; Touron-Poncet et al., 2014) rivers. Ephemeroptera are usually present in all stream types and benthic microhabitats within stream systems (Sowa, 1975), and show high morphological and ecological differentiation among genera (Domínguez et al., 2006). With such ecological characteristics, one may expect that variations in the biological trait combination of Ephemeroptera assemblages effectively account for ecosystem alteration.

The aim of this study was to assess the extent of shifts in the biological trait composition of Ephemeroptera assemblages along a gradient of disturbance associated with gold-mining in French Guiana (FG), East-Amazonia. Gold is the most significant mineral resource in the Guiana Shield (FG, Guyana and Surinam) (Hammond et al., 2007). After the prospection of large rivers, gold industries are now focusing on smaller inland streams (Cleary, 1990; Hammond et al., 2007). Small streams represent 80% of all running waters in FG and exhibit high ecological quality; some if not most of them have never been impacted by any human activity. Sediment discharges related to gold-mining activities are known to largely exceed those generated by other land-use changes, such as deforestation or road-building (Bruijnzeel, 1993; Krishnaswamy et al., 2006) and this type of disturbance certainly has harsh impact on the river biota in the Guiana shield where smalls streams naturally exhibit low levels of suspended materials (Hammond et al., 2007). In light of recent economic development, our ability to identify relevant reference conditions (e.g., community traits, ecological functions) and effectively rate ecosystem health will undoubtedly contribute to the success of future management actions. Assuming that environmental conditions strongly constraint Ephemeroptera assemblages (Hanquet et al., 2004), we hypothesized that (i) streams with similar habitat conditions host Ephemeroptera assemblages with similar combinations of traits, and (ii) anthropogenic disturbance generates broad shifts in ecological functions as species with certain traits are eliminated or replaced by species with other traits. In order to test these predictions, data on the Ephemeroptera assemblages were collected in 19 headwater streams of FG (abundance matrix for 35 genera), then five biological traits were described for the first time using a fuzzy-coding method (trait matrix). Matrix multiplication and a fuzzy-coding analysis were used to weight traits by taxa abundance, and to investigate the spatial distribution of trait combinations in relation to the extent of anthropogenic impacts generated by gold mining.

2. Materials and methods

2.1. Study area and sampling sites

This study was conducted in FG, East Amazonia, from October to December 2012. The climate is tropical moist with 3000–3400 mm of yearly precipitation mainly distributed over 280 days. There is a major drop in rainfall (dry season) between September and December and another shorter and more irregular dry period in March. The maximum monthly temperature averages 33.5 °C (32.1–35.8 °C), and the monthly minimum averages 20.3 °C (19.7–21 °C). The sampled streams had a water depth <1 m and a stream width <10 m, and were located in the upstream part of the river continuum. Larger streams and rivers were not considered in the study in order to focus on comparable ecosystems.

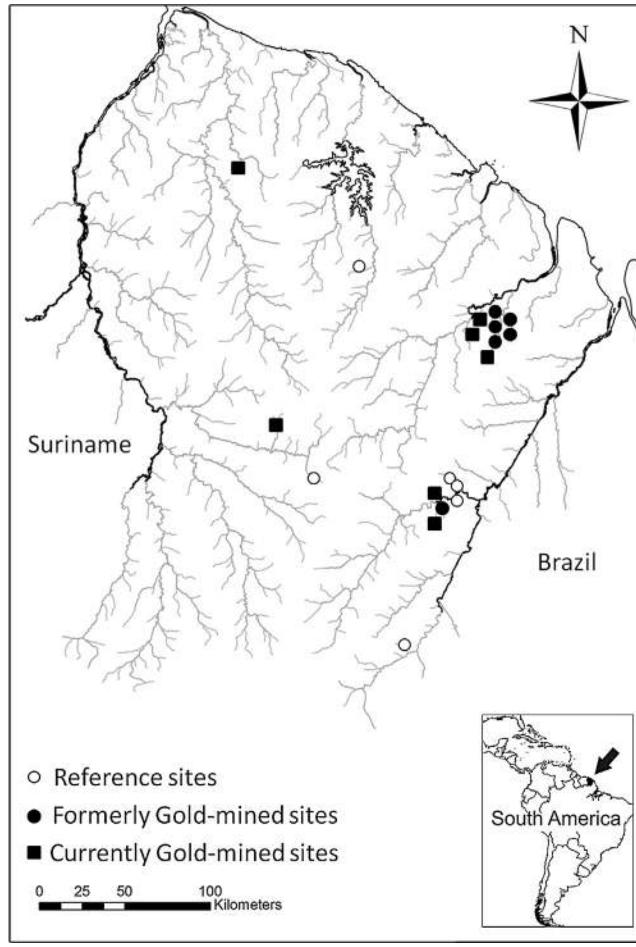

Fig. 1. Distribution of 19 sampling sites in French Guiana.

Nineteen sampling sites were sampled over 19 headwater streams belonging to FG's main river basins. Sites were mostly located on the northern part of FG, an area covered by dense rainforest and without road networks (Fig. 1). The sites were sampled during the dry season (September–December) in 2012. Six sites were defined as not subjected to anthropogenic impacts (Reference sites). Impacted sites were currently ($n = 7$) or formerly ($n = 6$) subjected to gold-mining. Gold mining activity had stopped 2 years before sampling at the formerly gold-mined sites. The mining activities considered in this study refer to so-called "illegal" mining, i.e., small-scale traditional (or artisanal) mining which occurs in most South American countries (Hammond et al., 2007). These illegal activities involve small groups of workers (around a hundred people) who settle on small and remote forest streams (Hinton et al., 2003).

2.2. Physical–chemical variables

The length of a site was defined as 10 times its width, and transects were established each 5 m along this length, for subsequent habitat measures. Stream flow (cm s^{-1}) and the percentage composition of organic and mineral substrate types were determined on a 1 m^2 area every meter along each transect. Mean stream flow at a site was the mean of all point measurements. The substrate types included: % litter, % submerged roots on the banks, % macrophytes, % woody debris, % sand (particle size <2 mm), % gravel (2–25 mm), % coarse substratum (>25 mm). Silt deposit being scarce naturally

Table 1
Physical–chemical characteristics of the three categories of sampling sites. Statistics are the results of Kruskal–Wallis test, comparisons made with values at the reference sites (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, ns: not significant).

Variables	Reference	Formerly gold-mined sites		Current gold-mined sites	
	Mean ± SD	Mean ± SD	Statistics	Mean ± SD	Statistics
Physical and water chemistry					
Temperature (°C)	24.96 ± 1.53	25.01 ± 1.16	ns	25.52 ± 2.13	ns
Conductivity (μs cm^{-1})	32.20 ± 24.69	35.10 ± 19.01	ns	45.10 ± 15.10	ns
pH	5.90 ± 2.59	6.01 ± 0.45	ns	6.04 ± 0.24	ns
Turbidity (NTU)	2.64 ± 1.85	6.96 ± 6.18	**	19.25 ± 9.38	***
Total Suspended Matters (mg L^{-1})	2.97 ± 0.79	6.30 ± 6.21	**	9.48 ± 11.78	**
Dissolved Oxygen (mg L^{-1})	6.97 ± 0.35	5.21 ± 1.45	**	6.53 ± 0.85	ns
Mean stream flow (cm s^{-1})	11.40 ± 9.67	6.35 ± 2.61	**	14.40 ± 2.74	*
Substrates					
% Silt	13.33 ± 6.83	45.50 ± 15.08	***	44.16 ± 14.97	***
% Sand	65 ± 15.49	29.50 ± 29.11	**	22.50 ± 17.81	***
% Gravel	21.67 ± 19.15	25 ± 21.44	ns	34.16 ± 19.61	*
% Woody debris	21.51 ± 10.65	18.31 ± 30.48	ns	25.31 ± 40.62	ns
% Macrophytes	11.22 ± 1.14	8.15 ± 3.21	ns	1.59 ± 3.11	***
% Submerged roots on the banks	35.13 ± 15.33	23.21 ± 24.45	*	11.25 ± 26.43	*
% Litter	22.50 ± 11.29	58.33 ± 25.81	***	55.83 ± 23.96	***

in FG streams, it was used to estimate the clogging of the river by gold mining.

We also measured chemical variables accounting for the impairment of stream ecosystem by human activities (Table 1). Four variables were directly measured in the field using probes: dissolved oxygen (WTW 3205®), turbidity (EUTECH®131), pH (WTW 3110®) and conductivity (WTW 3110®132). Water temperature (°C) was the mean of values given by all above-mentioned probes. Following standard methods (AFNOR, 2005), total suspended matters (mg L^{-1}) were measured in laboratory based on water samples taken at each site and immediately frozen.

Significant differences in physical–chemical conditions were compared among reference, formerly gold-mined and currently gold-mined sites using the Kruskal–Wallis (K–W) test followed by *posthoc* pairwise comparisons (Wilcoxon's test, hereafter *W* test) with a holm correction for multiple comparisons. Non-parametric tests were used because of small sample size and the heteroscedastic distribution of the assemblages.

2.3. Ephemeroptera sampling

Twelve sample units were taken at each site: 8 in organic substrates (roots, macrophytes, aquatic plants, litter, bryophytes) and 4 in mineral substrates (pebbles, gravels, sand, silt). Sample units in organic substrates consisted of intensive sweeping of a hand net (frame size = 46 × 23 cm; mesh size = 500 μm) through the substratum during 1 min over a 0.46 m × 1.5 m area (net width × 1.5 m). Sample units in mineral substrates were obtained by dragging a 5 cm-layer of sediment with the same net, over a 0.46 m × 1.5 m area. Prior to dragging, coarse particulates (pebbles) were brushed in front of the net, and then removed. The samples were preserved in the field in 4% (final concentration) formalin. Invertebrates were sorted in the laboratory and preserved in 70% ethanol. The individuals were sorted, identified and enumerated. The Ephemeroptera were identified to the genus level using the key of Domínguez et al. (2006). Owing to the strong phylogenetic conservatism in the study traits of Ephemeroptera at the family and genus level (Merritt and Cummins, 1996), we considered the genus level as relevant for subsequent analyses of trait combinations.

2.4. Ephemeroptera assemblages and biological traits

First, correspondence analysis (CA) was used to ordinate the sites according to the hellinger-transformed abundance matrix (Legendre and Gallagher, 2001) for the 35 genera, thus summarizing the variability of Ephemeroptera assemblages, and providing insights for the discussion of the subsequent biological traits analysis. The significance of the axes was determined at $p < 0.05$ by testing the eigenvalues of the inertia matrix. A permutational MANOVA (Anderson, 2001) was then used to evaluate differences in macroinvertebrate assemblage according to environmental characteristics and disturbance type. The significance of the test was given by *F*-tests based on sequential sums of squares from 1000 permutations of the data. As the order of non-orthogonal variables can have an effect on the outcomes of such method, the overall environmental descriptors of each microhabitat (habitat, substrates) and habitat (depth, width, velocity flow, pH) were first introduced in the analysis, and the mining-related variables (represented by turbidity and % silt) were the last considered. This approach was already implemented by Brosse et al. (2011) to take into account the pure effect of gold mining, in order to assess the effects of environmental characteristics and gold-mining intensity on fish communities of FG. Significant differences in genus richness and Shannon index were also evaluated among categories of disturbance, using K–W tests followed by *posthoc* pairwise comparisons (*W* test) with a holm correction for multiple comparisons.

The biological traits for each Ephemeroptera genus (Table 2, Appendix I) were obtained from the literature (Merritt and Cummins, 1996; Polegatto and Froehlich, 2003; Baptista et al., 2006; Domínguez et al., 2006; Tomanova et al., 2006, 2007), and the authors' observations of live and preserved specimens (e.g., locomotion, food acquisition, mouthparts). The biological traits examined were likely to respond to two major environmental selective forces in stream ecosystems: habitat heterogeneity/stability (locomotion, maximum body size, body form, gill form), and food resource (feeding group) (Poff et al., 2006). The categories for each trait were either ordinal or nominal. Categories used for the different traits are listed in Table 2. Information on the biological traits was then structured using a fuzzy-coding technique (Chevenet et al., 1994): scores ranged from '0', indicating 'no affinity' to '3', indicating 'high affinity' for a given trait category. This procedure allowed us to build a trait matrix. We then used multivariate analyses to evaluate whether the distribution of traits in Ephemeroptera assemblages could discriminate reference, formerly and current gold mined sites. The trait matrix was first multiplied by taxa abundance in each site. The site-trait array was log-transformed into relative abundance of each trait category in each site and further processed by Fuzzy Correspondence Analysis (FCA) (Chevenet et al., 1994). We then examined if the first FCA axis (FCA1), which

Table 2
Biological traits, categories (abbreviations as in Appendix I) and functional trends ("+": increase or "–": decrease) along the disturbance gradient (sediment addition and altered hydrology) resulting from significant correlations between the proportion of trait categories and the first Fuzzy Correspondence Analysis axis (FCA1) axis scores. p-Values are for Pearson's r tests: * $p < 0.05$, ** $p < 0.01$, ns: not significant.

Traits	Categories	FCA1	p-Value
Gill Form	Leaf like gills (LG)		ns
	Filamentous gills (FG)	–	**
	Plate like gills (PG)		ns
	Numerous Tracheal filament (TG)	+	**
	Operculate gills (OG)	+	*
Body Form	Streamlined		ns
	Flattened	+	**
	Cylindrical		ns
Maximal Size	<0.25 cm	–	*
	>0.25–0.5 cm		ns
	>0.5–0.7 cm		ns
	>0.7 cm	+	*
Feeding group	Brushers (Br)	–	**
	Scrapers (Scr)		ns
	Collector-Gatherers (CoGa)		ns
	Shredders (Shr)		ns
	Collector-Filterers (CoFi)	+	*
Mobility	Swimmers (Sw)	–	*
	Crawlers (Cr)		ns
	Epibenthic Burrowers (EpB)		ns
	Endobenthic Burrowers (EnB)	+	**

explained most of the variability in functional community composition, was related to the gold mining gradient. The relationships between FCA1 sites scores and the categories of impairments (reference, formerly gold-mined, currently gold-mined sites) were tested using a K–W test followed by *posthoc* pairwise comparisons (*W* test). Finally, Pearson's r coefficients were used to examine which trait categories were significantly correlated with FCA1 axis, in order to bring out the functional responses of taxa to anthropogenic perturbation.

All multivariate analyses and other statistical tests were implemented using the packages stats and ade4 (Chessel et al., 2004) in R 3.1.2 statistical software (R Core Team, 2014).

3. Results

3.1. Environmental variables

Physical–chemical variables (Table 1) differed significantly among the sites, according to the impairment type. Dissolved oxygen concentration and mean stream flow were significantly lower in formerly gold mined sites (K–W tests; $p = 0.0023$; $p = 0.0093$, respectively). Both currently and formerly gold-mined sites were characterized by higher turbidity and total suspended matters (K–W; formerly: $p = 0.0018$; $p = 0.0045$; currently: $p = 2.4\,E^{-5}$; $p = 0.0021$, respectively). The currently gold-mined sites had coarser substrates (gravels; K–W; $p = 0.0089$). The overall gold-mined sites (formerly and current) had more silt and less organic substrates (macrophytes and litter).

3.2. Responses of Ephemeroptera assemblages to gold-mining

The first and second CA axes described 19.01% and 15.32% of the total variability in Ephemeroptera assemblages, respectively (Fig. 2a). Such rather low explained variability is common when examining taxonomic assemblages (Fabrizi et al., 2010; Céréghino et al., 2012), while multivariate analyses of traits compositions provide much higher explanations of the total variability in the community structure (Dias et al., 2008; this study). A gradient of taxonomic structure matched a gradient of gold-mining activity along axis 2 (CA2) (Fig. 2b), and the distribution of site

Table 3
Results of the MANOVA performed on Ephemeroptera abundance data. The significance of the tests was checked using F-tests based on sequential sums of squares from 1000 permutations of the raw data (significant results are in bold).

Source of variation	MS	F	r^2	p
Micro-habitat scale				
% Sand	0.258	2.254	0.089	**0.020**
% Litter	0.270	2.360	0.094	**0.012**
% Roots	0.155	1.355	0.054	0.182
Habitat scale				
Width	0.152	1.330	0.053	0.233
Depth	0.099	0.865	0.034	0.507
pH	0.211	1.843	0.073	**0.048**
Stream Flow	0.145	1.262	0.050	0.267
Gold-mining				
Turbidity	0.243	2.123	0.084	**0.034**
% Silt	0.207	1.806	0.072	0.077
Residual	0.115		0.397	

coordinates along axis 2 differed significantly according to disturbance type (K–W; $p = 0.000171$—see Fig. 2b for pairwise-comparisons.). Axis 1 of the CA thus rather displayed environmental variability within each category of site, suggesting a wide variation in assemblage among sites within a category (see e.g., reference sites, Fig. 2a and b). Nevertheless, such within-category differences were due to uncommon (rare) genera, such as *Paramaka*, *Cryptonympha*, *Camelobaetidus*, *Terpides*, *Hydrosmilodon*. Some genera were, however, mainly found in gold-mined sites, namely *Hexagenia*, *Campusrus*, *Caenis*, *Apobaetids*, *Miroculis*, whereas other genera were associated with references sites (*Farrodes, Terpides, Rivudiva, Hagenulopsis*). A summary of the significance of the various effects of environmental parameters and gold-mining on the taxonomic structure of Ephemeroptera assemblages is given in Table 3. 61% of the total variance of the Ephemeroptera assemblages was explained by our measured variables (permutational MANOVA, total $R^2 = 0.603$). Differences in Ephemeroptera composition between sites were significantly explained by % litter, % sand and pH ($p = 0.020$, 0.012 and 0.049, respectively). After accounting for the effects of stream habitat and micro-habitat descriptors, gold-mining still explained 8.2% of the differences in assemblages between sites ($p = 0.034$). Moreover, significant differences across disturbance categories were apparent in terms of genus richness (K–W, $p = 0.02902$; Pairwise-Comparison Ref-Cug, $p = 0.01339$) and Shannon index (K–W, $p = 0.001679$, W, Ref-Cug, $p = 0.003405$; Fog-Cug, $p = 0.003361$) (Fig. 2c).

3.3. Responses of Ephemeroptera traits to gold mining

The first and second FCA axes described 57.85% and 22.29% of the total variability in trait composition of assemblages, respectively. A gradient of biological traits clearly matched a gradient of gold-mining activity along axis 1 (FCA1) (Fig. 3). Negative scores along FCA1 corresponded to reference and formerly gold-mined sites, whereas positive scores corresponded to the currently gold-mined ones. We found a significant difference of FCA1 coordinates between these categories of impairment (K–W; $p = 0.00149$—see Fig. 3b for pairwise-comparisons).

Some correlations between trait categories and FCA1 axis scores were statistically significant (Table 2). Within assemblages, the increasing proportions of large individuals (up to 0.7 cm), flattened bodies, endobenthic burrowers, collector-filterers and individuals with operculate gills or with tracheal filaments were correlated with the gold-mining gradient (Fig. 3a). Conversely, small individuals (<0.25 cm), brushers, swimmers and individuals with filamentous gills were preferentially associated to reference sites. We also noticed that formerly gold-mined sites were located on the middle area of the scatterplot, and that mid-sized organisms

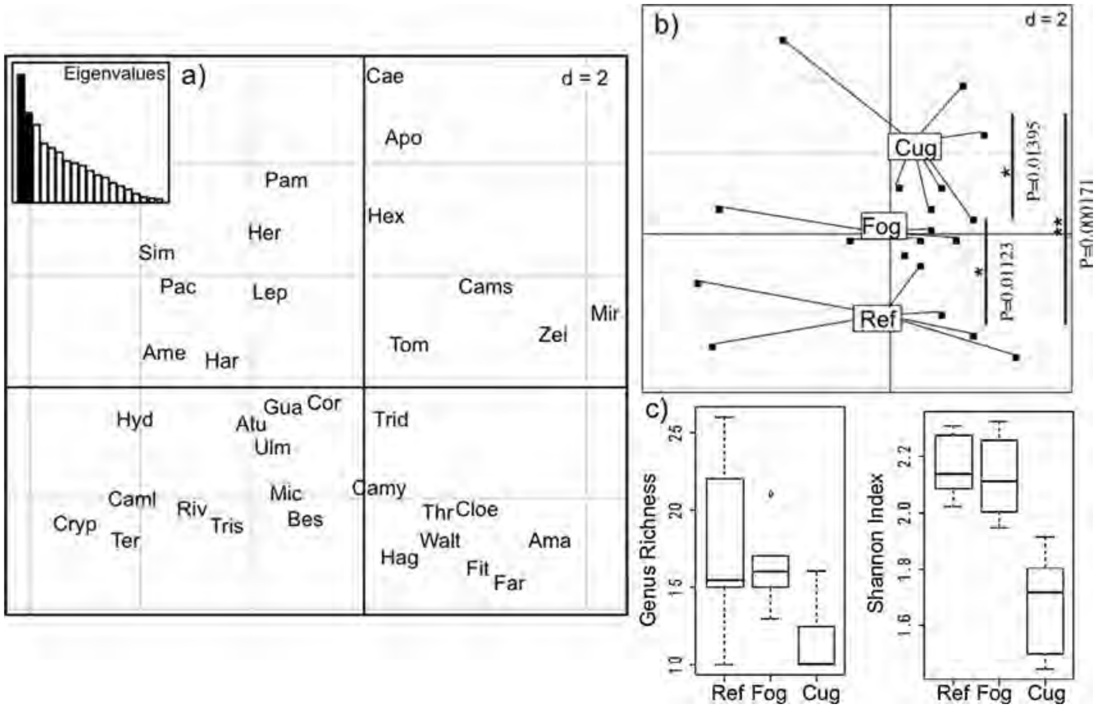

Fig. 2. Correspondence analysis (CA) of the taxonomic composition at the sampled sites. (a) Distribution of the genera in the first two factorial planes (See Appendix I for labels of genera), (b) distribution of sites in the first factorial plans. The mean position of the three categories is at the weighted average of corresponding sites. Sites were linked to their corresponding category, (c) genus richness and Shannon index of the different categories of disturbance (Ref: Reference sites; Fog: Formerly gold-mined sites; Cug: Currently gold-mined sites).

(0.5–0.7 cm), collector-gatherers, individuals with plate and leaf like-gills, crawlers, and epibenthic burrower seemed to be favored in these sites (Fig. 3a—Table 2).

4. Discussion

Biological traits of macroinvertebrates were successfully used in previous studies to reveal impacts of various types of disturbance, especially in the temperate zone where biological attributes of the benthic fauna are well documented (e.g., Dolédec et al., 1999, 2006; Usseglio-Polatera et al., 2000; Gayraud et al., 2003; Statzner et al., 2004). We know that biological traits (life history patterns, body size, etc.), species richness and numerical dominance do not compare among biogeographic regions, thus precluding the transposition of associations of biological traits and environmental conditions from temperate to tropical areas. Nevertheless, once biological traits are quantified for a given ecoregion (e.g., this study), we can take advantage of the strong phylogenetic conservatism of traits at the genus-family level to broaden their application beyond the local context used to generate them. In addition to biogeographic differences in traits, it is worth noting that the dominant economic activities that generate strong impacts upon river ecosystems do not fully compare between tropical (e.g., gold mining, timber for wood product) and temperate localities (e.g., intensive agriculture, industry, urbanization). Gold is the most significant mineral resource in the Guiana Shield (FG, Guyana and Surinam), and as such, has driven mining activity for centuries (Hammond et al., 2007). Ephemeroptera traits based on food acquisition, respiration and locomotion did well at detecting both past and current disturbance associated with gold-mining in headwaters. There was a clear shift of trait combinations from currently gold-mined sites to the formerly gold-mined ones, and then to reference sites. In light of our results, we can state that shifts in trait composition were related to changes in Ephemeroptera assemblage composition and richness.

Sediment addition is probably the structuring factor for assemblages in gold-mined sites. Several studies previously demonstrated that discharges from mining activities decrease species diversity and alter species composition (e.g., Beltman et al., 1999; Malmqvist and Hoffsten, 1999; Soucek et al., 2000; Tarras-Wahlberg et al., 2001; Yule et al., 2010). Vasconcelos and Melo (2008) documented the short-term impact of sediment release on tropical macroinvertebrates diversity. We found that higher proportions of endobenthic burrowers and collector-filterers were associated to gold-mined sites. In our study, this combination of traits corresponded to the so-called burrowing mayflies from the genera *Campsurus* (Polymitarcyidae) and *Hexagenia* (Ephemeridae), which are well adapted to deposited substrates (sand, silt). These Ephemeroptera inhabit U-shaped tunnels burrowed into clay and feed by resuspending the organic particles (Merritt and Cummins, 1996; Domínguez et al., 2006). We also noticed that individuals with leaf-like or filamentous gills were preferentially associated to undisturbed sites. Filamentous gills are considered as the most fragile organs in the Ephemeroptera order and may be linked to the absence of particles in the water column. Conversely, operculate gills (e.g., *Caenis*) act as protective covers thus conferring resistance to sediment increase. Gold-mining activities also lead to the loss of physical habitats such as macrophytes and mineral substrates. Habitats such as aquatic macrophytes provide direct (refuge habitat) or indirect (support for the development of algae and biofilm that constitute food) resource supplies (Allan and Castillo, 2007). In reference sites, we mainly found herbivorous taxa (brushers and scrapers) feeding over the surface of coarse mineral particles. This result corroborates former studies in neotropical areas which showed that sediment release due to gold-mining clog the river bottom (Mol and Ouboter, 2004). Higher sediment loads into the watercourse lead to the decline of primary producers, and subsequently, the herbivores (e.g., the Baetidae in our study) that depend on them (Parkhill and Gulliver, 2002; Suren et al., 2005; Tudesque et al., 2012).

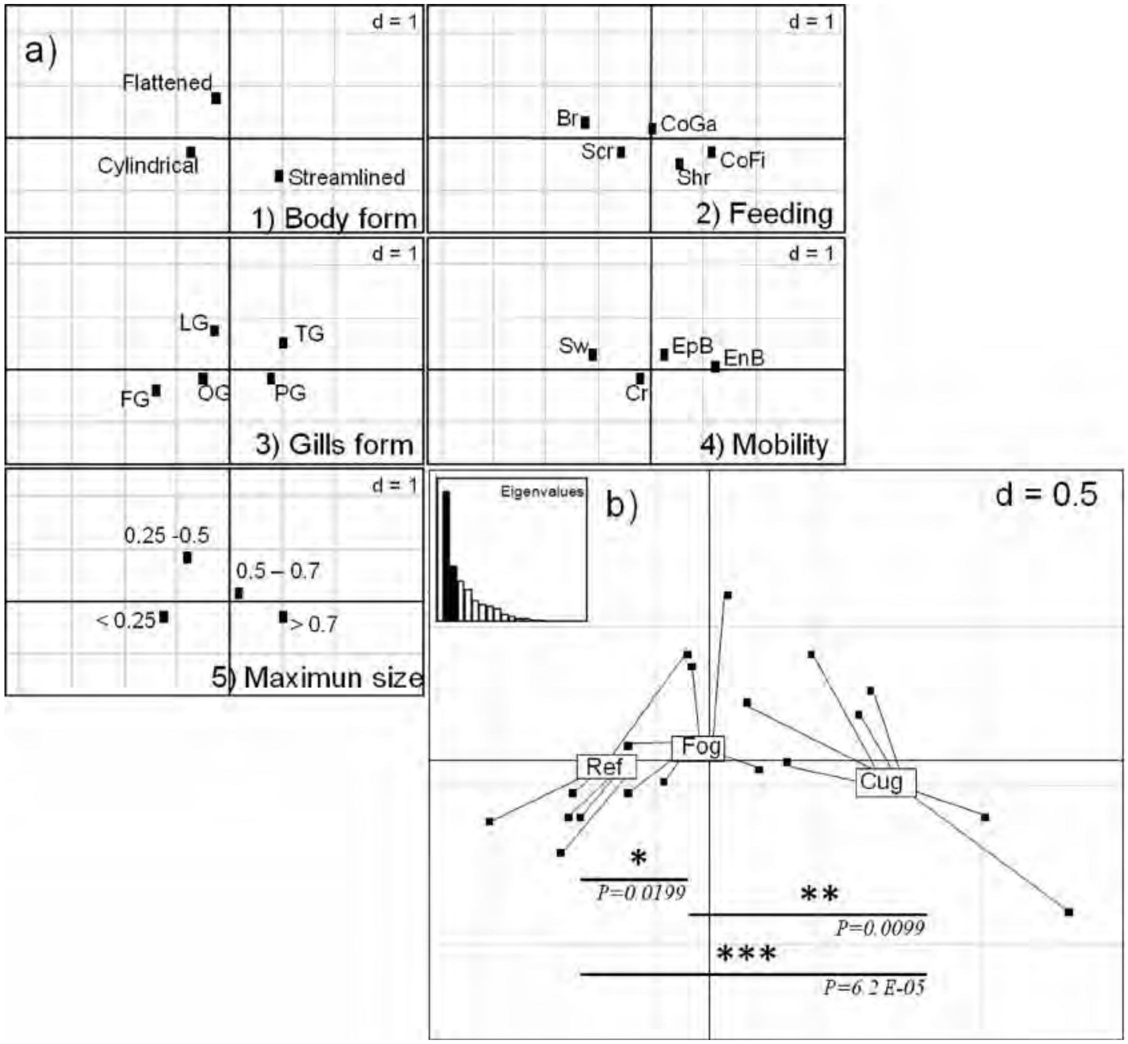

Fig. 3. Fuzzy correspondence analysis (FCA) on functional composition in sampled sites. (a) Distribution of categories of the five biological traits in the first factorial plan (see Table 2 for labels of categories), (b) distribution of sites in the first factorial plan. The mean position of the three categories was located at the weighted average of corresponding sites. Sites were linked to their corresponding category (Ref: Reference; Fog: formerly gold-mined; Cug: current gold-mined).

The above-discussed results suggest a strong relationship between habitat conditions and Ephemeroptera traits. However, some differences in Ephemeroptera diversity also accounted for variations in local environmental conditions. Regardless of human-induced perturbation, variations in % sand, % litter and pH were the most structuring factors. Where allochthonous inputs in the form of coarse organic matter are important (e.g., litter), it is expected that shredders should be the numerically dominant non-predators. At sites where autochthonous energy is more important, scrapers should have greater numerical importance. Most of our sites were located in forested areas, with a more or less dense canopy cover by overhanging tress. They thus exhibited variable accumulations of leaf litter over the bottom. Many of the dominant Ephemeroptera were associated with leaf debris, notably *Campylocia* (filterer-colllector), *Farrodes* (scraper) and *Waltzohyphius* (collector), thus explaining the importance of the variable % litter. Sandy substrates are typical microhabitats of the burrowing mayflies (see above). Ephemeroptera diversity is known to be adversely affected by low pH values (Bell, 1971; Sutcliffe and Carrick, 1973; Rowe et al., 1988; Courtney and Clements, 2000; O'Halloran et al., 2008). The genera *Camelobaetidius*, *Hermanella* and *Thraulodes* were only found in remote reference sites with higher pH (>5.8). These results suggest that neotropical Ephemeroptera assemblages were partly shaped by deterministic processes (Lepori and Malmqvist, 2009). Finally, it is likely that the stochastic occurence of rare taxa like *Paramaka, Terpides, Hydrosmilodon* contributes to between-site variability in assemblage diversity in reference and impacted sites. It is worth noting that no significant decline in genus richness and Shannon entropy was observed in formerly gold-mined sites 2 years after abandonment, compared to the reference sites, but the taxonomic and trait composition of communities changed at these sites. Both traits and taxa compositions thus have the potential to reveal the persisting effect of the harsh physical modifications that once occurred with mining (e.g., deforestation, channelization, creation of artificial pools) on current benthic assemblages. Similarly, Brosse et al. (2011) found an incomplete resilience of fish assemblages after the abandonment of sites in FG, in areas where mining activities had stopped for one year. However, we note that biological metrics such as richness or Shannon index, commonly used as basic indicators of ecosystem impairment, or even to design multimetric indices in the neotropics (Baptista et al., 2007; Helson and Williams, 2013; Villamarin et al., 2013; Touron-Poncet et al., 2014) were inefficient at detecting the "ghost of past disturbance". Assuming that current multimetric indices developed for neotropical streams do well at quantifying shifts in taxonomic structure, the inclusion of trait-related metrics could lead to relevant improvements of these biological assessment tools. Moreover, recolonization by Ephemeroptera can occur through the oviposition of aerial adults, upstream movement, downstream drift or movement upward from the hyporeic zone (Williams and Hynes, 1976). The contribution from these various sources of colonists will vary with species and situations (Townsend and Hildrew, 1976). Further works on the quantification of new traits, notably those related to life history patterns (e.g., number of generations per year), reproduction (e.g., number of eggs per female adult) and dispersal ability (e.g., drifting behavior, flight abilities) should therefore help to evaluate more precisely the effect of human disturbances on the ecological integrity of neotropical streams (Brittain, 1991; Dolédec et al., 1999, 2006).

5. Conclusion

Although the biological traits approach to biomonitoring has many benefits, its use in neotropical streams is still in its infancy. Beyond the need to document species traits through studies of individual and population biology, there will be a need to determine which traits best respond to which type of human impacts. For instance, Ephemeroptera assemblages may not be sensitive to a full range of potential disturbances encountered in eastern Amazonia, although they do well at detecting gold-mining impacts in this area. Traits of sensitive groups such as the Ephemeroptera could also be considered in multimetric indices that comply with the EU Water Framework Directive guidelines, while such biomonitoring tools are progressively set up and implemented in overseas region of Europe (Touron-Poncet et al., 2014; Dedieu et al. submitted). Lastly, our published trait profiles of Ephemeroptera will hopefully contribute to future trait approaches intended to compare ecological functions in pristine and impacted streams in nearby neotropical areas (Appendix I).

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Appendix A. Appendix I: Fuzzy-coded traits of 35 mayfly genera for five functional traits (see text). Abbreviations of trait categories as in Table 2.

Family	Genus	Code	Feeding group					Maximal body size				Mobility				Gill form					Body form		
			Scr	Shr	Br	CoGa	CoFi	<0.25	>0.25–0.5	>0.5–0.7	>0.7	Sw	Cr	EpB	EnB	LG	FG	OG	TG	PG	Flatt	Stream	Cylin
Baetidae	*Americabaetis*	Ame	3	0	1	0	0	0	0	3	0	3	2	0	0	0	0	0	0	3	0	0	3
	Apobaetis	Apo	2	2	0	1	0	0	3	0	0	2	2	0	0	0	0	0	0	3	0	0	3
	Aturbina	Atu	3	0	1	0	0	0	3	0	0	2	2	0	0	0	0	0	0	3	0	0	3
	Camelopbaetidus	Caml	3	0	0	0	0	2	2	2	0	3	2	1	0	0	0	0	0	3	0	0	3
	Cloedes	Cloe	3	0	0	2	0	3	0	0	0	2	3	0	0	0	0	0	0	3	0	0	3
	Cryptonympha	Cryp	3	0	1	0	0	0	2	2	0	2	2	1	0	0	0	0	0	3	0	0	3
	Guajirolus	Gua	3	0	0	0	0	0	3	0	0	3	2	0	0	0	0	0	0	3	0	0	3
	Harpagobaetis	Har	3	0	1	0	0	0	2	2	0	3	2	0	0	0	0	0	0	3	0	0	3
	Paracloedes	Pac	3	0	0	0	0	0	3	0	0	2	3	0	0	0	0	0	0	3	0	0	3
	Rivudiva	Riv	0	0	3	2	0	0	2	3	0	2	3	0	0	0	0	0	0	3	0	0	3
	Tomedontus	Tom	0	0	3	2	0	0	2	2	0	0	1	3	0	0	0	0	0	3	0	0	3
	Waltzoyphius	Walt	3	0	0	0	0	0	3	0	0	2	3	0	0	0	0	0	3	0	0	3	0
	Zelusia	Zel	3	0	0	0	0	0	3	0	0	2	3	0	0	0	0	0	3	0	0	3	0
Caenidae	*Caenis*	Cae	2	0	0	3	0	0	3	0	0	0	3	1	0	0	0	0	3	0	3	0	0
Coryphoridae	*Coryphorus*	Cor	1	0	0	2	0	0	2	2	0	0	2	3	0	0	0	3	3	0	3	0	0
Ephemeridae	*Hexagenia*	Hex	0	0	0	3	1	0	0	0	3	0	0	2	3	0	0	0	0	3	0	3	0
Euthyplociidae	*Campylocia*	Camy	0	2	0	1	3	0	0	0	3	0	1	3	2	0	0	0	3	0	0	3	0
Leptohyphidae	*Amanahyphes*	Ama	1	0	0	3	0	0	3	0	0	0	3	1	0	0	0	3	0	3	3	0	0
	Leptohyphes	Lep	0	1	0	3	0	2	2	0	0	0	2	2	0	0	0	3	0	3	3	0	0
	Tricorythopsis	Tris	1	0	0	3	0	2	2	0	0	0	3	0	0	0	0	3	0	3	3	0	0
	Tricorytodes	Trid	1	0	0	3	0	0	3	0	0	0	3	2	0	0	0	3	0	3	3	0	0
Leptophlebiidae	*Bessierus*	Bes	0	0	3	2	0	0	3	0	0	0	1	2	0	0	3	0	0	0	3	0	0
	Farrodes	Far	0	0	3	2	0	0	3	0	0	3	2	0	0	0	3	0	0	0	3	0	0
	Fittkaulus	Fit	3	0	1	0	0	0	2	2	0	3	2	0	0	3	0	0	0	0	3	0	0
	Hagenulopsis	Hag	0	0	3	2	0	0	3	0	0	0	1	2	0	3	0	0	0	0	3	0	0
	Hermanella	Her	3	0	1	0	0	0	3	2	0	3	2	0	0	3	0	0	0	0	3	0	0
	Hydrosmilodon	Hyd	3	0	0	1	0	0	2	2	0	0	1	3	0	3	0	0	0	0	3	0	0
	Microphlebia	Mic	0	0	3	2	0	0	3	0	0	2	2	0	0	3	0	0	0	0	3	0	0
	Miroculis	Mir	0	0	3	2	0	0	0	3	0	2	2	0	0	3	0	0	0	0	3	0	0
	Paramaka	Pam	3	0	0	0	0	0	2	2	0	2	3	0	0	3	0	0	0	0	3	0	0
	Simothraulopsis	Sim	1	0	3	2	0	0	2	0	0	2	2	0	0	3	2	0	0	0	3	0	0
	Terpides	Ter	0	0	2	3	0	0	0	3	0	0	2	3	0	3	0	0	0	0	3	0	0
	Thraulodes	Thr	0	0	3	2	0	0	0	3	0	0	1	3	0	3	0	0	0	0	3	0	0
	Ulmeritus	Ulm	0	0	3	2	0	0	2	2	0	1	1	2	0	0	0	0	3	0	3	0	0
Polymitarcyidae	*Campsurus*	Cams	0	0	0	2	1	0	0	2	2	0	1	3	2	0	0	0	3	0	0	3	0

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Résumé : La Directive Cadre Européenne sur l'Eau (DCE) a imposé à chaque état membre l'objectif d'obtenir une amélioration de la qualité des eaux de surface, pour d'atteindre le bon état écologique à l'horizon 2015. Cette directive implique non seulement d'évaluer le degré de pollution mais aussi de définir l'état écologique des milieux par rapport à un référentiel (se rapprochant de l'état « naturel »). Cela a conduit à définir une typologie des rivières basées sur les «conditions de référence». Bien que géographiquement éloignés du continent, les DOM font partie intégrante de l'Union européenne et sont soumis aux mêmes objectifs et obligations en termes de politiques environnementales. La Guyane française (GF) est une région d'outre-mer de la France située sur la côte nord-est de l'Amérique du Sud. La forêt primaire guyanaise reste l'une des moins touchés du monde, cependant, les exploitations aurifères et sylvicoles ont de fortes répercussions sur les milieux aquatiques. Les petits cours d'eau (profondeur inférieur à 1 m et da largeur inférieur à 10 m) représentent 70-80% du réseau hydrographique de la GF. La plupart des petits cours d'eau sont situés dans les zones boisées et présentent haute qualité écologique et certains, sinon la plupart d'entre eux, n'ont jamais été touchés par une activité humaine. L'objectif principal de cette thèse était de concevoir un indice capable d'évaluer la qualité écologique des petits cours d'eau de la GF. Nous avons d'abord analysé l'effet des perturbations humaines sur la qualité physicochimique des écosystèmes. Puis, nous avons établi la première typologie de la structure des communautés benthiques en fonction des conditions environnementales. L'identification de ces sous-régions écologique et des conditions de références nous a permis de construire un indice multimétrique. Enfin, une approche fonctionnelle basée sur les Ephémères a été utilisée afin de fournir de nouvelles informations sur l'écologie de cet ordre. Nous avons démontré que les variables physiques relatives au fond des cours d'eau et les matières en suspension différencient les sites orpaillés, tandis que les concentrations en nutriments ne sont pas significativement modifiées par les impacts humains. La biotypologie a montré que les sites caractérisés par des communautés d'invertébrés sont regroupées en deux sous-régions principales: la plaine alluviale côtière et le plateau des Guyanes. Les changements dans la composition de la communauté, et à un degré moindre la richesse taxonomique au sein de chaque sous-région ont révélé des impacts écologiques liés à l'exploitation aurifère et forestière. Parmi les mesures décrivant la structure des communautés (métriques) calculées à partir des deux sous-ensembles de sites de références et impactés, nous avons sélectionné les paramètres présentant le meilleur compromis entre forte efficacité de la discrimination, faible spécificité, faible redondance et une grande stabilité dans les conditions de référence. L'IBMG est composé de deux métriques taxonomiques de richesse, deux métriques basées sur l'abondance, une métrique relative au trait biologique et un indice de diversité. Les résultats sur les communautés d'Ephémère ont mis en évidence le rôle important de cet ordre pour la bioévaluation de petits cours d'eau soumis à l'exploitation aurifère. Des recherches futures fondamentales sur la taxonomie et l'écologie de cet ordre pourront permettre d'améliorer l'outil d'évaluation biologique actuel. Enfin, ce travail à souligné le fait que les petites masses d'eaux de tête de bassin de Guyane abritent potentiellement une faune riche et spécifique.

Mots clés : Bioindication ; invertébrés benthiques ; écologie des communautés ; néotropical ; Directive Cadre Européenne ; écologie aquatique.